The Thesis Committee for Kimberly Marie Bittler
Certifies that this is the approved version of the following thesis:

The Ecology of Blue Crab (*Callinectes sapidus*) Megalopae in the Mission-Aransas Estuary, Texas: Salinity, Settlement, and Transport

APPROVED BY
SUPERVISING COMMITTEE:

Supervisor:

Edward J. Buskey

G. Joan Holt

Richard B. Forward

The Ecology of Blue Crab (*Callinectes sapidus*) Megalopae in the Mission-Aransas Estuary, Texas: Salinity, Settlement, and Transport

by

Kimberly Marie Bittler, B.A.

Thesis

Presented to the Faculty of the Graduate School of

The University of Texas at Austin

in Partial Fulfillment

of the Requirements

for the Degree of

Master of Science in Marine Science

The University of Texas at Austin

December 2013

Dedication

This thesis is dedicated to my family and friends. Thank you for supporting me!

Acknowledgements

I want to thank my supervisor, Dr. Ed Buskey, and my committee members, Dr. Joan Holt and Dr. Richard Forward, for their crucial support and guidance. Thanks to Deepak Adhikari and Dr. Brad Gemmell for their contributions to the tomographic PIV characterization of flows, Dr. Jim Welch for mailing me megalopae from North Carolina, Abbie Sherwin and Josh Pullin for help counting samples and overnight sampling, and Dr. Andrew Esbaugh, Dr. Lee Fuiman, and Dr. Ben Walthers for lending me equipment. I want to thank Dr. Zack Darnell for his advice and guidance, and Dr. Pablo Munguia. Thanks to Cammie Hyatt for her constant support, and Tracy Harvey for being an awesome lab "sister" and sharing an office with me. I especially want to thank Dr. Lindsay Scheef for her constant guidance, for teaching me how to use R, and for camping out on the pier for overnight sampling every single night we tried.

Abstract

The Ecology of Blue Crab (*Callinectes sapidus*) Megalopae in the Mission-Aransas Estuary, Texas: Salinity, Settlement, and Transport

Kimberly Marie Bittler, MSMarineSci

The University of Texas at Austin, 2013

Supervisor: Edward J. Buskey

Blue crabs are a widely distributed estuarine species with broad economic and ecological importance. Several studies have linked blue crabs to freshwater inflows, but the precise nature of this link is still uncertain, as blue crabs have a complex life cycle that utilizes both marine and estuarine environments. One potential link between blue crabs and freshwater inflows is during recruitment, when megalopae developing offshore return to estuaries before molting into juvenile crabs. Megalopae swim during the flood tide to ensure delivery into and farther up estuaries. The behaviors regulating selective tidal stream transport (STST) on the flood tide were originally studied in North Carolina in an estuary with regular freshwater inflows and a strong salinity gradient. The model of STST was re-examined in the Mission-Aransas, an estuary with episodic freshwater inflows and salinity gradients ranging from normal estuarine conditions to hypersaline during droughts. The behavioral responses of megalopae to a range of rates of salinity increase were tested, and then modeled onto rates of salinity change observed in the field

to determine the theoretical ecological consequences of STST for blue crab populations in the Mission-Aransas Estuary.

To validate the ecological trends predicted by the behavioral model of STST, a simple, long-term data set reflecting changes in megalopae abundance is needed. Hog's hair collectors are a simple and widely used method of quantifying abundance of brachyuran megalopae, including blue crabs. However, the efficiency of hog's hair collectors in sampling for megalopae is unknown. Several studies have reported poor correlations between settlement on hog's hair collectors, transport, and abundance of megalopae in the plankton due to disparate temporal scales and potentially turbulence-driven decoupling. Each of these issues were addressed in field and flume experiments, which were used to develop a model for interpreting settlement on hog's hair collectors in terms of transport and planktonic abundance.

Table of Contents

List of Tables

List of Figures

Chapter 1: Freshwater Inflows and Blue Crabs: The Influence of Salinity on Selective Tidal Stream Transport

ABSTRACT

Freshwater inflows are crucial for the function of estuaries, but can become limited due to drought and human use. Several studies have linked blue crab populations to freshwater inflows, suggesting that inflows are critical for an early life history event such as recruitment during selective tidal stream transport as megalopae, and lowered predation and increased growth rates as juveniles. This study focused on selective tidal stream transport (STST) by re-examining the behavioral responses of *Callinectes sapidus* megalopae to various rates of salinity increase. The goal was to determine if the behaviors regulating selective tidal stream transport (STST) provide a plausible link between blue crab populations and freshwater inflows. The results of the behavior experiments showed that Texas megalopae have more variable responses to rates of salinity change than a study conducted in North Carolina, and that this variance lies between cohorts rather than within cohorts or experimental treatments. This study found that the rate of salinity increase that elicits the maximum behavioral response is half the rate reported in a previous study. This higher sensitivity is adaptive for transport into the drought prone Mission-Aransas Estuary when an estuarine gradient is present, but not for drought conditions, as export out of the estuary is possible during drought periods due to increasing salinity on the ebb tide. A simple behavior-driven flux model shows that transport of megalopae out of the estuary during drought is possible, and may lead to recruitment failures in the absence of other recruitment mechanisms.

INTRODUCTION

Freshwater inflows supply nutrients to estuaries and maintain low salinity nursery areas that are critical to the productivity and function of estuarine systems (Longley 1994, Powell et al. 2002). Despite the importance of maintaining flows into estuaries, freshwater resources are becoming limited as coastal development and the demand for water increases. The decrease in freshwater reaching estuaries is projected to continue, and to be exacerbated by the effects of an increasingly arid climate in Texas (Montagna et al. 2011). Freshwater inflows may be most important to Texas estuarine ecosystems during drought years, when salinities in some river-fed bays such as Copano Bay in the Mission-Aransas estuary, can approach or even rise above the salinity of sea water (CDMO 2013). Understanding the ecological effects of salinity fluctuations and variable freshwater inflows is crucial to the effective management of estuaries in changing environments.

Blue crabs (*Callinectes sapidus*) are an important species in Gulf of Mexico estuaries, both as a commercial fishery and because of their trophic role in estuarine ecosystems (Hoeinghaus & Davis 2007). Several studies have linked freshwater inflows to blue crab abundances and commercial landings in Florida (Wilber 1993), North Carolina (Posey et al. 2005), Louisiana (Guillory 2000) and Texas (More 1969). Wilber (1993) found that a one to two year lag in freshwater inflows in Apalachicola Bay Florida could explain temporal variability in commercial blue crab landings, and suggested that nutrient supplies or low salinity refuges from predators as juveniles were responsible for the link. Blue crabs have a complex life cycle, broad salinity tolerances, and the ability to move large distances within estuaries throughout their life (Aguliar et al. 2005). These factors make studying the specific mechanisms that regulate the connection between salinity, freshwater inflows, and abundance difficult.

Spawning females migrate to higher salinities at the mouths of estuaries (Carr et al. 2004, Aguliar et al. 2005) to release multiple clutches of larvae known as zoea, which require full ocean salinity to develop (Darnell et al. 2009). The planktonic zoea live in the ocean for 31-49 days before molting into a megalopal stage (Costlow and Bookhout 1959). The megalopae are advected towards estuary mouths by wind driven currents (Epifanio 1995) and move farther up estuary with behavioral adaptations that take advantage of hydrologic movements, such as tides (Forward et al. 2003). These behavioral responses are triggered by physical factors such as increasing salinity and turbulence (Welch and Forward 2001), and possibly by chemical cues associated with estuaries (Forward and Rittschof 1994). The Texas coast has nearly continuous barrier islands separating the Gulf of Mexico from the estuaries, with widely separated narrow passes. These limited passes into the estuaries may make larval recruitment an especially important component of blue crab population dynamics on the South Texas coast.

Once within an estuary, megalopae settle on vegetation then quickly molt into first instar juveniles. Juvenile crabs are an important food source for many estuarine species, such as red drum (Scharf & Schlicht 2000) and the endangered whooping cranes (Chavez-Ramirez 1996). Juvenile crabs are found in higher densities in salt marshes and other vegetated habitats that may provide better refuge from predation relative to unvegetated bottom (Minello et al. 2003). In estuaries where vegetated habitats are limited, low salinity areas may provide an alternate nursery habitat with lower predation risk and enhanced food supply (Posey et al. 2005). Therefore, there are two life stages when freshwater inflows are likely to be particularly important to blue crabs: when recruiting back to estuaries as megalopae (Welch and Forward 2001), and when susceptible to heavy predation as juveniles (Posey et al. 2005). This study will focus on

the recruitment stage, specifically the behavioral responses of megalopae that ensure transport into estuaries.

Megalopae transport and salinity cues

The behaviors that govern transport via tides are well understood from studies performed on the US Atlantic coast. Transport is generally limited to the night, as the chemical signature of estuarine waters induces photoinhibition of megalopae activity during daylight, and megalopae only actively swim at night when in the estuarine plume. Welch and Forward (2001) experimentally demonstrated a mechanism for the transport of blue crab megalopae into Atlantic coast estuaries known as selective tidal-stream transport (abbreviated STST; later reviewed by Forward et al. 2003). Their model proposed that megalopae utilize nocturnal flood tides to move up estuaries, and avoid being transported back out to sea on the ebb tide through a series of responses to changes in salinity and turbulence:

(1) Megalopae swim up into the water column in response to increasing salinity and pressure

(2) Megalopae remain swimming in response to high levels of turbulence

(3) Megalopae descend when turbulence declines

(4) Megalopae are inhibited from rising again with the ebb tide by decreasing salinity and pressure

While this model is plausible for estuaries on the Atlantic coast that have larger tidal ranges and more consistent freshwater inflows, several issues arise when applying this behavior-response model to transport in systems like the Mission-Aransas Estuary in Texas. In the Gulf of Mexico, tidal ranges are relatively small (Smith 1977). These weaker tides may result in rates of pressure and salinity change too low to stimulate a

swimming response (Tankersley et al. 1995), and move smaller volumes of water than more extreme tides observed on the Atlantic coast. Tidal currents alone may not be enough to transport blue crab megalopae into Texas estuaries, and a model of planktonic larval transport for the area has suggested that wind forcing is a more important process driving transport of larval fish (Brown et al. 2005). The Mission-Aransas and other Texas estuaries also experience more extreme drought conditions than estuaries on the Atlantic Coast. For instance, in dry years some estuaries can become more saline than the ocean when freshwater inputs fall below the rates of evaporation. During these conditions the increases in salinity that cue upward swimming in blue crab megalopae would occur during the ebb tide rather than the flood tide, potentially transporting the megalopae away from estuarine nursery habitats. It is also possible that the changes in salinity during drought conditions may not be of a great enough magnitude to initiate an upward swimming response from the megalopae at all.

Tankersley et al. (1995) performed experiments that explored the effect of different rates of salinity change on the vertical distribution of North Carolina blue crab megalopae (*Callinectes sapidus*) under dark conditions in seawater. The megalopae and water used in their study were collected from a more typical estuary with reliable freshwater inflows and a strong salinity gradient. The variable conditions in the Mission-Aransas Estuary make this location an ideal place to test whether salinity's role in STST varies between regions with differing estuarine characteristics. Our findings are applicable to other estuaries that may also have more extreme salinity patterns, especially as freshwater inflows become more limited. This study will examine the salinity response of megalopae collected from the Mission-Aransas Estuary, and examine the effects that these behaviors may have on recruitment with the following hypotheses:

(1) Megalopae swim upward in response to changes in salinity when held in offshore water (independent of any ambient estuarine chemical signal) and this behavior does not vary between cohorts.

(2) Megalopae swim upwards in response to the same rates of salinity change as megalopae tested by Tankersley et al. (1995) when held in ambient flood tide water (and are exposed to any estuarine chemical cues present in this flood tide water)

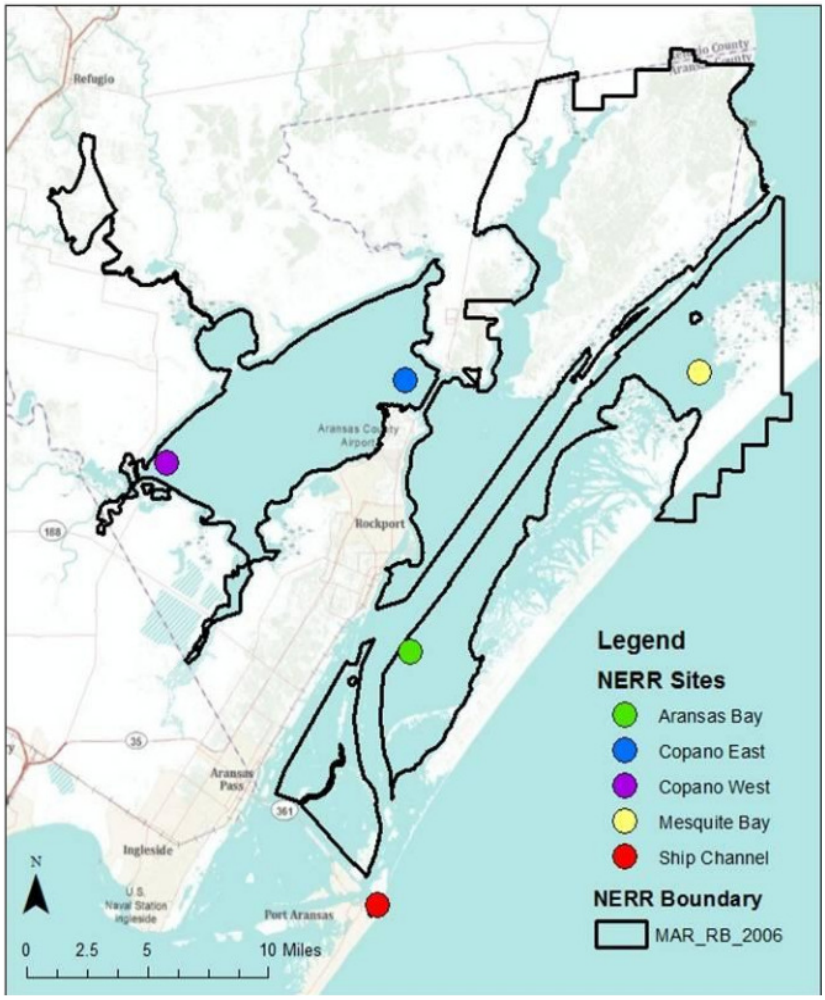
(3) There is no difference in the vertical swimming behavior of megalopae collected in Texas versus North Carolina when exposed to identical flood tide water and salinity cues.

METHODS

Study site

The Mission-Aransas is a bar built estuary in south Texas that has relatively low impacts from development (NOAA 2006). The small watershed is predominantly influenced by coastal weather patterns, with freshwater inputs draining into the Mission and Aransas rivers. These two rivers flow into Copano Bay, which is connected to Aransas Bay. Aransas Bay exchanges water with San Antonio Bay to the north through the Gulf Intracoastal Waterway and with the Gulf of Mexico to the south through the Port Aransas Ship Channel. The Ship Channel is the primary inlet not only for the Mission-Aransas Estuary, but also for Corpus Christi Bay and the hypersaline Laguna Madre, resulting in complex salinity patterns.

Figure 1.1: Map of the Mission-Aransas National Estuarine Research Reserve (MANERR) boundaries and System Wide Monitoring Program Stations (SWMP)



The Mission-Aransas Estuary is within the Mission-Aransas National Estuarine Research Reserve (MANERR) established in 2007 to carry out long term environmental research that informs effective management decisions. The MANERR maintains five long term System Wide Monitoring Program (SWMP) stations throughout the estuary that continuously monitor temperature, salinity, dissolved oxygen, pH, turbidity, and water level at 15 minute intervals with YSI 6600 EDS data sondes suspended 0.5 m from the bottom of the water column, which is typically 5 m deep (Figure 1.1). This dataset is

a valuable source of long term, seasonal, and tidal trends experienced in the estuary. We used the data collected at these stations to calculate the range of salinity change rates that megalopae recruiting into the estuary might experience during both normal and drought years. These rates were compared to those that Tankersley et al. (1995) found to induce upward swimming behavior in megalopae in North Carolina.

Behavior experiments

A series of three experiments was conducted to explore the behavioral responses of locally caught blue crab megalopae to changes in salinity. Following Tankersley et al. (1995), we built a vertical 18 x 6 x 6 cm Lucite flow-through chamber in which the ambient salinity could be gradually changed by pumping in water through a small opening at the base. To confine the megalope to the center portion of the chamber where their response could be video recorded (Cohu 6500 camera with Sony Digital 8 Video Walkman), the column was divided into three sections by removable 83 µm mesh secured onto two 6 cm diameter PVC pipes at the top and bottom of the chamber. A circular acetate sheath of 6 cm diameter was wrapped around the PVC to keep megalopae out of the corners of the chamber while reducing the effects of parallax when filming. All experiments were conducted in a dark room illuminated by dim red light, and the experimental chamber was back-illuminated with infrared LEDs (wavelength > 850 nm). All megalopae were allowed to dark adapt for at least 10 minutes before any experiment began and were allowed to adjust to the experimental chamber for 3-5 minutes before video recording. Experiments were conducted at room temperature (~24º C).

Salinity within the chamber was manipulated by pumping water into the bottom section with a multi-speed peristaltic pump, and both increases and decreases in salinity entered the chamber from below. A stir bar at the bottom of the chamber ensured mixing

of the water and uniform flow, and displaced water was allowed to spill through a drain at the top of the chamber to ensure water pressure would remain constant. Salinity change rates were calibrated according to the chamber dimensions and pump speed, and were verified by measuring both the initial and final salinities in the chamber.

Megalopae were collected from the University of Texas research pier in the Port Aransas Ship Channel (27º 50' N, 97º 3' W) during nocturnal rising tides with a 0.5 m diameter 500 µm mesh bongo net. Megalopae were sorted into 10 cm diameter glass bowls, fed freshly hatched *Artemia franciscana* nauplii, and held at 20 ºC under a 12:12 light dark cycle until experiments were conducted. All experiments were conducted within 3 days of collection, and only intermolt megalopae (i.e., megalopae not showing signs of preparing to molt) were used. Megalopae were tested in groups of 25-30, and were transferred into water used for experiments at least one hour before experiments were conducted. For the first two experiments, megalopae were used for one treatment only, and were not re-used in other treatments.

Ocean salinity experiment

The first experiment was designed to assess the innate variability in behaviors between megalopae cohorts, as our second experiment would require us to use megalopae collected on different dates. Megalopae collected on dates at least one week apart were considered to be independent cohorts. We subjected megalopae from three different cohorts to a change in salinity when held in ocean water (33.3 ppt, collected in the Gulf of Mexico, 30 miles offshore). Since megalopae behavior may be altered by chemical signals (Forward and Rittschof 1994), salinity was manipulated using deionized water and salt extracted from the offshore seawater by evaporation, and the same offshore water was used for each cohort. Salinity was either increased or decreased by 0.6 ppt at a rate of

$1.0x10^{-3}$ ppt s^{-1}, a rate within the range observed at the study site (see Table 1.1) and known to stimulate the maximum upward swimming response in megalopae from North Carolina (Tankersley et al. 1995). The response was measured as the number of megalopae actively swimming in the top 2/3 of the viewing chamber post-stimulus compared to the number of megalopae actively swimming before the stimulus was applied, following the procedure of Tankersley et al. (1995). Controls were conducted on each group of megalopae by circulating water at the same flow rate (0.12 mL s^{-1}) without any changes in salinity, and allowing the megalopae to recover without any stimulus for 3-4 minutes before the experimental stimulus was applied. For each of the three cohorts (Table 1.2), the experiment was conducted with three replicates per treatment. Data were arctangent transformed to produce normal distribution, and were analyzed with an ANOVA mixed model, which estimates variation due to cohort, treatment (increase or decrease in salinity), and treatment within a cohort. All data analysis and statistics were conducted in R (R Core Team 2013).

Flood tide rates experiment

The second experiment examined the response of megalopae to a set of positive salinity change rates: 0 (control), $2.5x10^{-4}$, $5x10^{-4}$, $1x10^{-3}$, $2\ x10^{-3}$, and $4\ x10^{-3}$ ppt s^{-1}. These rates encompass the range observed during flood tide at the study site and are within the range tested by Tankersley et al. (1995). Megalopae were tested in the same flood tide water from which they were collected to capture the water chemistry conditions experienced by the megalopae as they were actively recruiting into the estuary through the Ship Channel. Salinity was increased with salts derived from ocean water. During each collection date, every rate including the control was tested once without replicates following a complete block design. The response was tested by comparing each

mean to the control with Student's t-tests. Five separate cohorts were tested, collected at least one week apart (Table 1.3) in order to capture the variability present in independent cohorts accurately.

Megalopae origin comparison experiment

The third experiment explicitly compared the responses of megalopae collected in the Mission-Aransas Estuary to megalopae collected from North Carolina. North Carolina megalopae were collected from the plankton during the rising tide the night of July 29, 2013 at the entrance to the Newport River Estuary in Beaufort, North Carolina (34º 43' N, 76º 40' W), placed in a seawater saturated paper towel, and mailed overnight to Texas in a cooler. The megalopae arrived on July 31, 2013 and were transferred to offshore water (37 ppt, collected in the Gulf of Mexico, 30 miles offshore, May 2013), fed freshly hatched *Artemia* nauplii, and held in an incubator overnight at 20°C 12:12 h L:D cycle before any experiments were conducted. Texas megalopae were caught on nocturnal flood tides from the University of Texas research pier on August 1 and 2, 2013. Between experiments, all megalopae were maintained in offshore water in 5 cm diameter petri dishes, fed freshly hatched *Artemia* nauplii, and held in a 20 ºC incubator (12:12 l:d cycle). Flood tide water (37.1 ppt) was collected on August 2, 2013 from the University of Texas research pier, and was used for all experiments. To ensure water chemistry was as consistent as possible between experiments, the flood tide water was held in a dark refrigerator in a polycarbonate carboy, and allowed to return to room temperature before use in experiments.

Megalopae from Texas and North Carolina were divided into three separate groups of 25 megalopae. Each group was tested for every rate of salinity change ($2.5x10^{-4}$, $5x10^{-4}$, $1x10^{-3}$, $2\ x10^{-3}$ ppt s^{-1}), as well as a negative control (0 salinity change) and a

positive control (repeat $5x10^{-4}$ ppt s^{-1} salinity change) to ensure repeated testing did not affect the behavior of the megalopae. The order of the treatments was randomized for each group, and the megalopae tested were allowed to recover in the experimental flood tide water for at least two hours between treatments. The arctangent transformed data were analyzed using a repeated-measures ANOVA, where the separate groups were treated as true replicates to avoid pseudoreplication.

Field modeling component

We developed a simple qualitative model to explore how the behavioral responses measured in the above experiments might affect the recruitment of megalopae into the Mission-Aransas Estuary. This model predicts what proportion of megalopae present on a given night will be transported into or out of the estuary (positive or negative P_{flux}, respectively) based on the maximum proportion that would respond to the range of salinity changes on the flood tide that night (P_{flood}) minus the maximum proportion that would respond on the ebb tide that night (P_{ebb}), such that:

$$P_{flux} = P_{flood} - P_{ebb}$$

This approach is justified since the salinity response of megalopae is short, lasting only minutes, and once megalopae have ascended into the water column they are carried within a parcel of water and will not experience further changes in salinity (Welch and Forward 2001).

Actual rates of salinity change in the field were estimated based on data from two of the MANERR SWMP stations: the Ship Channel site, which is the major pass that megalopae developing offshore must be transported through to reach the estuary, and the

Aransas Bay site, which is further up the estuary (although megalopae are rarely collected on hog's hair collectors this far up estuary (unpublished data)). Salinity is measured every 15 minutes at these stations by YSI 6600 V2 Multiparameter Water Quality Sondes, and has been collected at a depth 0.5 m from the bottom since August 2007 at the Ship Channel site, and since April 2007 at the Aransas Bay site. For this analysis, we assumed that the water column was well mixed, which is likely given the rapid current speeds and high turbulence of water moving through the channel under most conditions.

The rate of salinity change was calculated per 15 minute interval, and smoothed within an hour window to lessen the impact signal noise from conductivity sensor and random processes (such as eddy shedding), and all rates over 2.22×10^{-3} ppt s^{-1} were excluded from the analysis following standard quality control protocols of the NERR system (any change in salinity over 2 ppt within the 15 minute sampling period is rejected by the CDMO (2013) data management protocols, equivalent to a calculated 2.22×10^{-3} ppt s^{-1} rate of salinity change over the sample period). The responses of megalopae to rates of salinity change measured in this study and from the Tankersley et al. (1995) study were estimated for each rate of salinity increase in the field with linear interpolation. Values measured during the day (between sunrise and sunset) were excluded from the model, as most transport of megalopae occurs at night (Forward et al. 2003)

The direction of the tide at the Ship Channel site was determined using velocity data collected by a Nortek Aquadopp Acoustic Current Profiler at 0-2 m depth from June 2010 to December 2012. While regression models predicting the tidal velocity were successful at the Ship Channel site (r^2=0.66), the model was not precise enough to include in our behavioral transport model (RMS=0.25) as the most extreme salinity changes happened at the lowest current speeds at the beginning and end of tides. Tidal

direction at the Aransas Bay site from June 2010 to December 2012 was estimated from the astronomical tidal harmonic (TCOON, 2013) and validated with velocity data collected by OkeanoLog Seahorse tilt current meters from December 2012 to February 2013. The model explains 55.7 % of the variance in current velocities under low wind conditions (>7 m s-1), and was used to predict tidal direction for the transport model.

Estimates of P_{flux} were integrated from June 2010 to December 2012 to estimate net flux of megalopae over the time period for which current velocity estimates were available. 95% confidence intervals for mean net flux estimates were calculated by bootstrapping (1,000 iterations). Alternate models in which salinity values were left unsmoothed or were averaged by hour were examined to assess model's sensitivity to treatment of the salinity data.

This model does not account for potential changes in water pressure, source-sink dynamics, or water chemistry, so flux calculations represent an estimate of net movements of megalopae due to salinity regimes alone. While increasing pressure is a reliable cue of the flood tide in some estuaries, our analysis indicates that increasing pressure is not a reliable indicator of the flood tide in the Mission-Aransas Estuary due to high levels of noise (e.g. from waves) and a small tidal amplitude (<1 m). Furthermore, any rates of pressure change generated by astronomical tides in the Mission-Aransas are far below the rates found to elicit a response in megalopae from the east coast (Tankersley et a. 1995), justifying the exclusion of pressure as a strong transport mechanism in blue crab megalopae into the Mission-Aransas estuary. Source-sink processes may drive transport into the estuary, with a supply of megalopae freshly developed offshore, and a sink for megalopae settling within the estuary. Estuaries also have unique chemical signatures that megalopae may use to refine selective tidal stream

transport behaviors. However, neither of these transport mechanisms is addressed in the scope of this paper.

RESULTS

Rates of salinity change in the Mission-Aransas Estuary

The average rate of salinity increase across all SWMP sites in the NERR during the study period is 3.07×10^{-4} ppt s^{-1}, with mean rates of increase from 3.48×10^{-4} ppt s^{-1} at Copano West to 2.78×10^{-4} ppt s^{-1} at Copano East. The overall average increase is slightly below the minimum rate of change that Tankersley et al. (1995) found sufficient to stimulate a significant response (5.53×10^{-4} ppt s^{-1}) and results in a suitable absolute change to stimulate megalopae response over 15 minutes (0.4 ppt per 15 minutes). During wet years, such as in summer of 2010, the mean change in salinity is higher at most sites compared to drought years such as the summer of 2011 (Table 1.1).

Table 1.1: Average rates of salinity changes (ppt s^{-1}) observed across MANERR SWMP sites from June 2010 to December 2012. Only increases in salinity are compared between drought (August 2011-January 2012) and normal (August 2010-January 2011) conditions.

Site	Increase	Decrease	Drought	Normal
Aransas Bay	2.98×10^{-4}	-3.05×10^{-4}	2.79×10^{-4}	3.29×10^{-4}
Copano East	2.78×10^{-4}	-2.82×10^{-4}	2.10×10^{-4}	3.17×10^{-4}
Copano West	3.48×10^{-4}	-3.47×10^{-4}	4.59×10^{-4}	2.58×10^{-4}
Ship Channel	2.89×10^{-4}	-2.92×10^{-4}	2.14×10^{-4}	4.11×10^{-4}
Overall	3.07×10^{-4}	-3.10×10^{-4}	2.65×10^{-4}	2.91×10^{-4}

Behavior experiments

Ocean salinity experiment

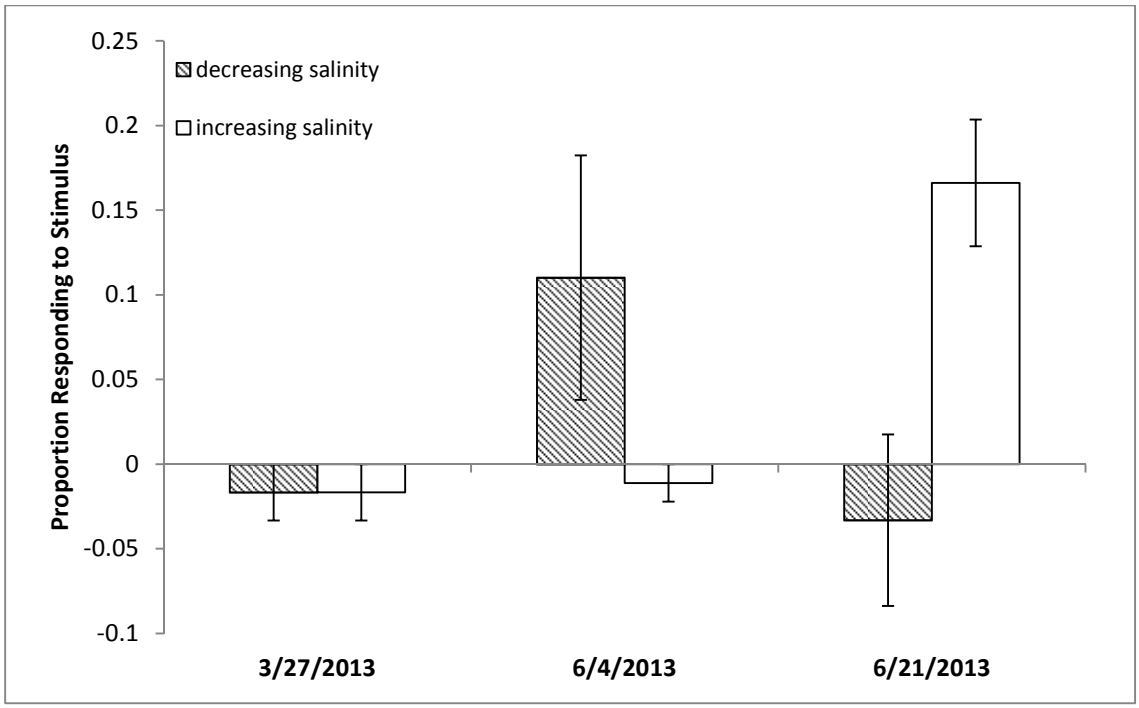
There was high variability in the salinity responses of the three different cohorts (collection dates on Table 1.2), despite holding experimental conditions constant (Figure

1.2). The first cohort had almost no response to either an increase or decrease in salinity, while the second cohort appeared to have a non-significant positive response to decreasing salinity (rather than the expected positive response to increasing salinity). Only the third cohort showed the expected response to salinity, with a positive response to increasing salinity, and a slightly negative response to decreasing salinity. There were non-significant main effects of cohort (F=2.32 *df*=2 p=0.14) and of changing salinity (F=.6008 *df*=1 p=0.4533), however changing salinity nested within cohort was significant (F=7.88 *df*=2 p=0.0065). There was a significant difference between the increase and decrease in salinity for the third cohort only (p=0.043, Tukey HSD test).

Table 1.2: Field conditions when megalopae were captured during flood tide for the ocean salinity experiment, measured by SWMP station.

Date	**Time**	**Salinity**	**dS/dt**
3/27/2013	6:15 AM	33	0
6/4/2013	10:00 PM	28.4	$-1x10^{-3}$
6/21/2013	12:00 AM	28.8	$5.6x10^{-4}$

Figure 1.2: Net response to an increase or decrease in salinity ($1.0x10^{-3}$ ppt s^{-1}) when tested in offshore water with no estuarine cues across three cohorts with standard error (n=3). A positive response indicates an increase in the proportion of megalopae in the upper 2/3 of the experimental chamber, while a negative response indicates a decrease.



Flood tide rates experiment

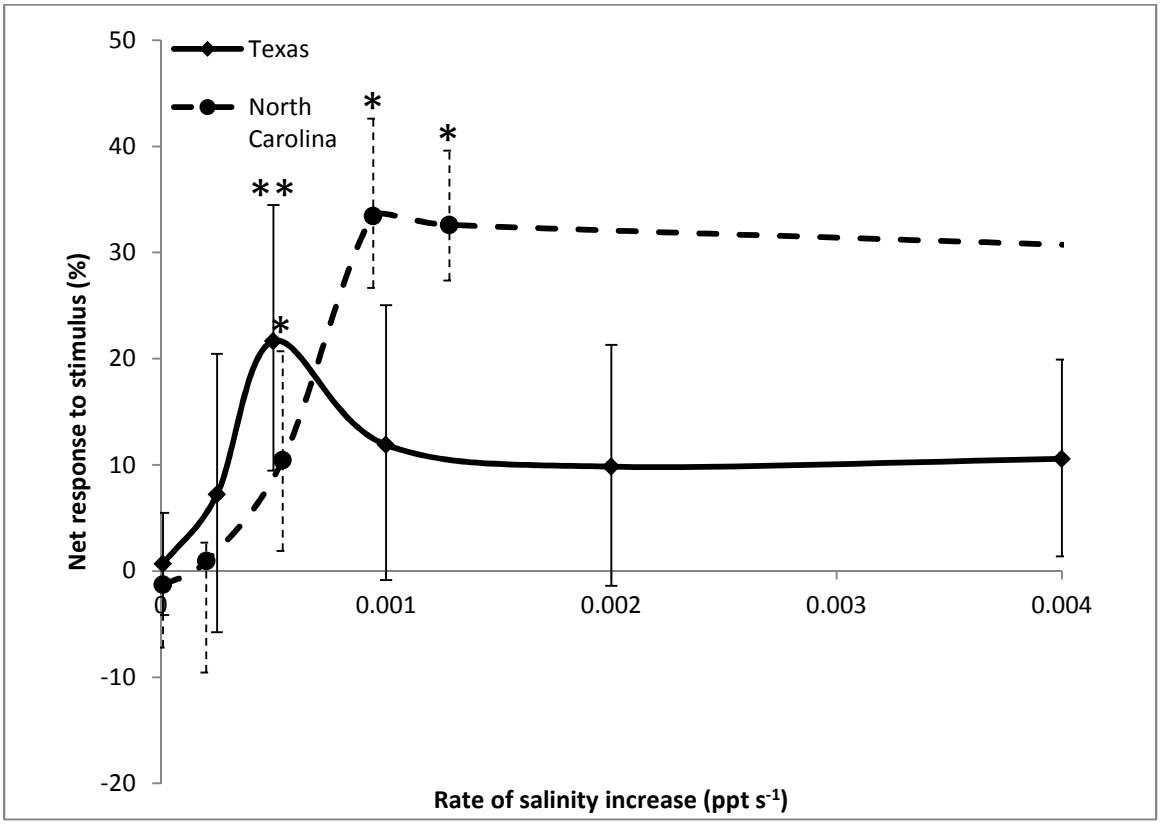
The responses to a relative rate of salinity increase were tested in 5 separate cohorts (Table 1.3) of megalopae across a geometric series of rates from $2.5x10^{-4}$ to $4.0x10^{-3}$ ppt s^{-1}. The variability observed in the ocean salinity experiment carried over into the rates experiment, where both the magnitude of the net response (a positive net response indicating an increase in the proportion of megalopae in the upper 2/3 of chamber, and negative response indicating a decrease), and the rate of salinity change that elicited the maximum response varied between $2.5x10^{-4}$ and $2.0x10^{-3}$ ppt s^{-1} among cohorts. When all cohorts were considered together, there was a distinct maximum net response of 21.6 % (95 % confidence interval: 9.5 – 34.5%) at $5x10^{-4}$ ppt s^{-1}. The

response to more rapid rates of salinity change was dampened and not significantly different from controls (Figure 1.3). The maximum response at $5x10^{-4}$ ppt s^{-1} was the only response that was significantly different from the controls ($p=0.02$), which were not significantly different from a net response of zero ($p=0.799$), justifying that any response was due to the changing salinity, and not artifacts of the experimental procedure or random movement.

Table 1.3: Field conditions during megalopae capture and water collection for the flood tide rates experiment, taken from SWMP station. dS/dt represents the rate of salinity change in ppt s^{-1}. Turb indicates turbidity, measured in Nephelometric Turbidity Units (NTU).

Megalopae				**Water**				
Date	**Time**	**Salinity (ppt)**	**dS/dt (ppt s^{-1})**	**Date**	**Time**	**Salinity (ppt)**	**Turb (NTU)**	**pH**
5/21/2013	5:00 AM	33.1	0	5/21/2013	12:00 PM	34.7	5	8.1
6/5/2013	12:00 AM	28.4	$-1x10^{-3}$	6/5/2013	11:00 AM	25	2	8.1
6/20/2013	12:00 AM	35.8	$-5.6x10^{-4}$	6/20/2013	11:40 AM	35.8	8	7.9
7/9/2013	6:00 AM	36.4	0	7/9/2013	11:10 AM	35.9	4	7.8
8/17/2013	2:15 AM	36.6	0	8/17/2013	11:00 AM	36.7	1	8

Figure 1.3: Net response (±95% confidence interval) to various rates of salinity increase in flood tide water compared between this study and a study conducted in North Carolina (data from Tankersley et al. 1995). Double asterisks indicate net response significantly greater than control for megalopae from Texas (solid line), and single asterisk indicates net response significantly greater than control for North Carolina megalopae (Tankersley et al. 1995, dotted line).



Megalopae origin comparison experiment

The response to various rates of salinity increase was directly compared between megalopae collected from Texas and from North Carolina using a repeated measures experimental design (Figure 1.4, conditions during collection in Table 1.4). The main effect of megalopae origin was significant (F=6.193 *df* = 1 p=0.03), as were the main effects of rate of salinity change when repeated measures were accounted for (F=7.287 *df*

= 4 p=0.0009). The effect of testing group (F=1.441 *df*=5 p=0.246) and all interactions were not significant and explained very little variability. Texas megalopae had higher responses than the North Carolina megalopae to the same rates of change, but the rate-response curve was of the same shape with a maximum response at $1x10^{-3}$ ppt s^{-1} for both groups, as shown by the non-significant interaction of origin and rate. Negative controls for both origins were not significantly different from zero (p=0.236 for NC, 0.692 for TX), and the negative controls for each origin were not significantly different from each other (p=0.300), indicating that the divergence noted between the responses of North Carolina megalopae and Texas megalopae is likely happening as salinities are increased and is a sensory driven behavior process. The comparison between repetitive stimulation at 5x10-4 ppt s-1 increase in salinity (the positive control for repeated measures) were not significantly different for either Texas (p=0.3636) or North Carolina (p=0.1938) megalopae, which indicates treatment effects were short-lived and validates the repeated measures experimental design.

Table 1.4: Field conditions when megalopae were captured for the comparison experiment, and the conditions flood tide water was collected in.

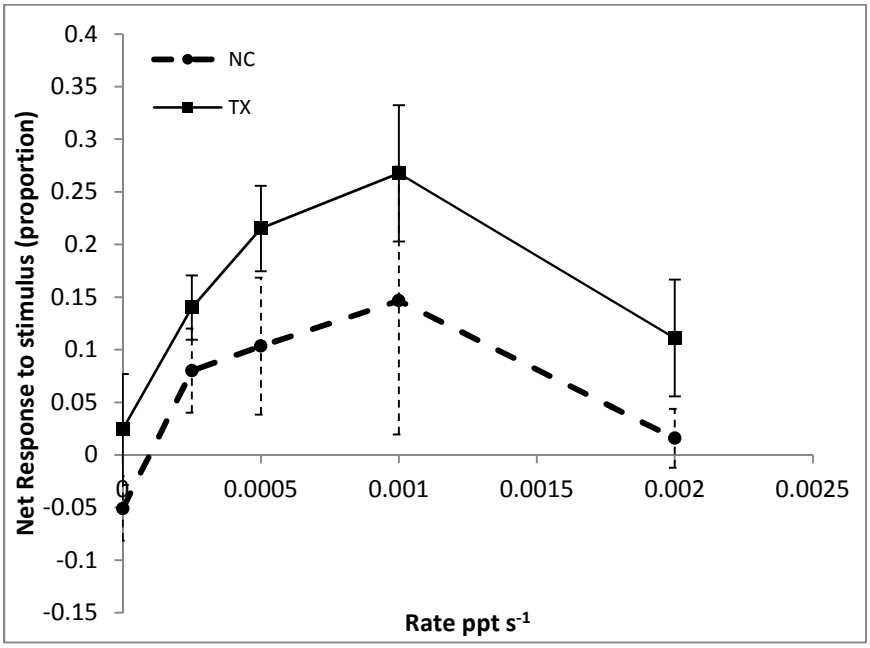
Megalopae

Date	Time	Salinity	dS/dt	
7/29/2013	-	32-35	-	NC
8/1-2/2013	5:00 AM	36.9	0	TX

Water

Date	Time	Salinity	Turb	pH
8/2/2013	5:20 AM	37.1	3	8

Figure 1.4: Net Response (±95% confidence interval) to various rates of salinity increase in flood tide water: a comparison between megalopae collected in North Carolina and megalopae collected in Texas.



Model results

Salinity at the ship channel ranged from 12.6 to 41.3 ppt over the 2 year study period, and from 8.8 to 41.5 ppt at the within-estuary Aransas bay site, generally increasing during drought and decreasing during wet years (Figure 1.5). Daily maximum rates of salinity change at the Ship Channel site ranged between -4.4×10^{-4} and 2.0×10^{-3} ppt s^{-1} on the ebb tide with a median at 3.3×10^{-4} ppt s^{-1}, while the flood tide had a lower median at 1.1×10^{-4} ppt s^{-1} and a less extreme range from -4.4×10^{-4} to 1.8×10^{-3} ppt s^{-1}. While the frequency of a low daily maximum rate of salinity increase ($0\text{-}2.5 \times 10^{-4}$ ppt s^{-1}) was higher on the flood tide at the Ship Channel, rates of change higher than 2.5×10^{-4} ppt s^{-1} occurred more frequently on the ebb tide (Figure 1.6).

Figure 1.5: Average daily salinity across all sites at the MANERR from June 2010 to December 2012. Asterisks indicate rain events (>40.0 mm day^{-1}), black bar indicates drought period used in model, and gray bar indicates flood period used in model. (site sc=Ship Channel, ab=Aransas Bay, ce=Copano East cw=Copano West, listed in order of closest to Gulf to farthest up estuary. For site locations, see Figure 1.1).

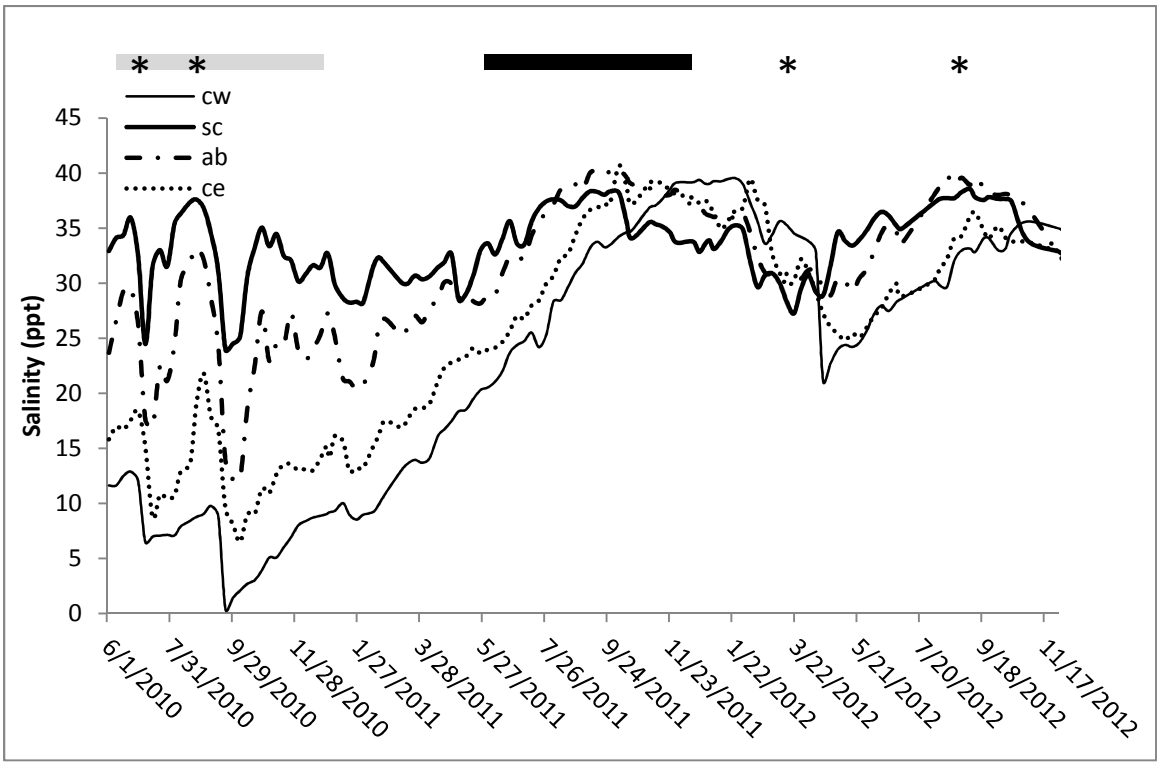
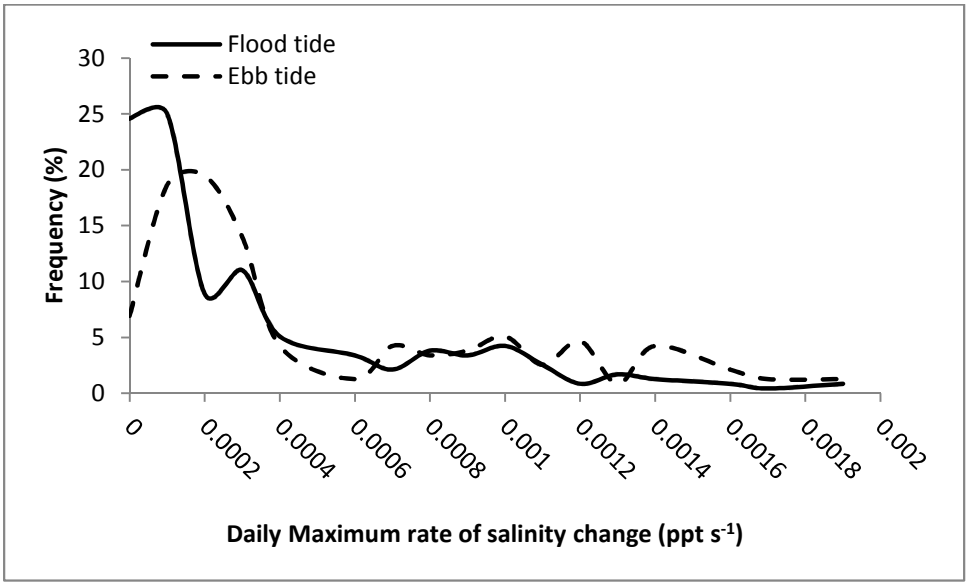
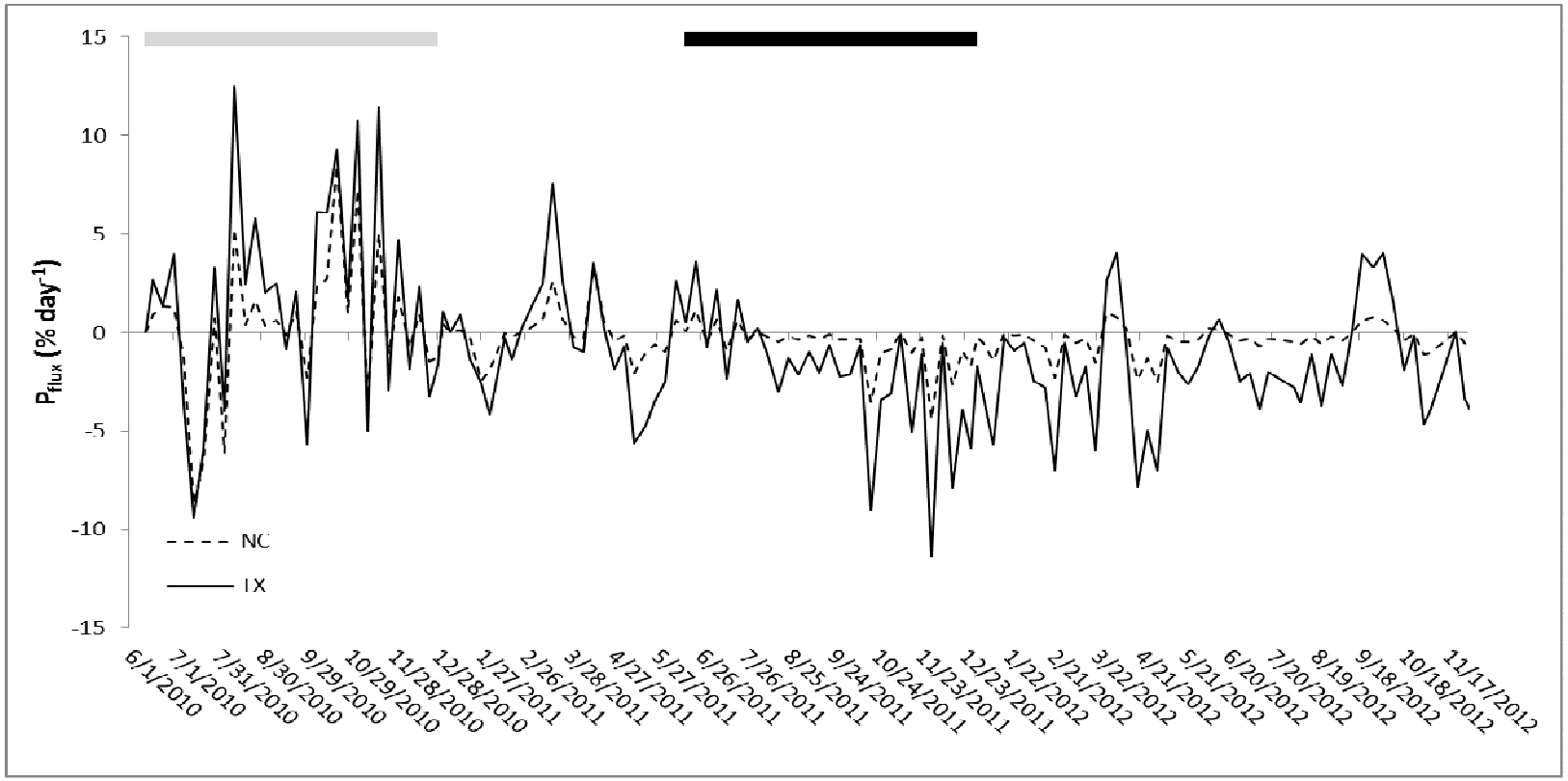


Figure 1.6: Percent frequency of daily maximum rate of salinity change observed at the Ship Channel MANERR site on the ebb and flood tides from June 2010 to December 2012. Only increases in salinity are included.



The model using rates of change of salinity measured in the Ship Channel and the response curves from the experiment series (rates in flood tide water) predicted an overall slight negative flux of Texas megalopae out of the estuary over the 2.5 year time series (-0.84 % day^{-1}, 95% confidence interval: -1.22, -0.45). However, positive net flux of megalopae into the estuary using salinity signals is substantial (4.11 % day^{-1}) when a strong estuarine gradient is present such as the fall of 2010 (Figure 1.7), as is the flux out of the estuary during drought conditions. The negative flux out of the estuary is due to increases in salinity on the ebb tide, and during these drought periods an average of 4.5 % of megalopae are expected to be transported out of the estuary per day.

Figure 1.7: Results of behavioral transport model, predicting net flux as % per day, averaged by week from June 2010 to December 2012 through the Aransas Ship Channel. Positive P_{flux} indicates import of megalopae into the estuary while a negative value indicates export of megalopae out of the estuary. Gray bar indicates a wet time period, while black bar indicates period of drought.

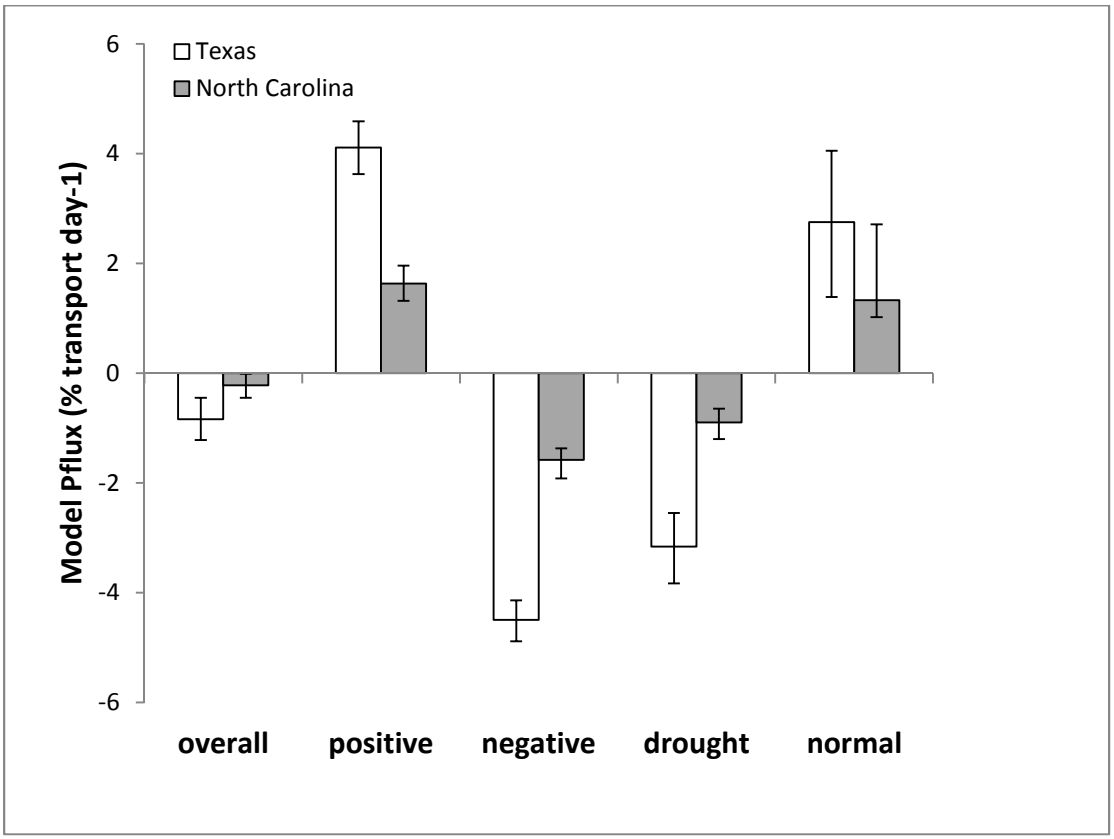


When the model was constructed at the Ship Channel using the rate-response curve of megalopae tested from North Carolina by Tankersley et al. (1995), fewer megalopae are expected to be transported out of the estuary over the whole time series (-0.22% day $^{-1}$, 95 % confidence interval: -0.45, -0.01). When the model results are separated into dates when negative P_{flux} was predicted and positive P_{flux} was predicted, it becomes apparent that the lesser export of the Tankersley-based model is due to an overall lower response (Table 1.5, Figure 1.8). This lower response is due to the higher threshold rate of salinity change needed for North Carolina megalopae that is rarely reached in the Mission-Aransas Estuary. Twice as many Texas megalopae are transported into the estuary (4.11% day^{-1}, 95% confidence interval: 3.63, 4.59) compared to the Tankersley-based model (1.62% day^{-1}, 95% confidence interval: 1.32, 1.96). P_{flux} follows climate trends, with the differences in model performance still favoring transport of the Texas megalopae over the Tankersley model in a wet period from July 2010 to January 2011. Any advantage gained by the Texas model compared to the Tankersley model is lost during periods of drought such as July 2011 to January 2012 when over twice as many megalopae are predicted to be exported out of the estuary in the Texas model compared to the Tankersley model. Most of the 2.5 year study period was during a severe drought (2009, 2011-present), so the advantage of the Texas model during normal conditions was not reflected in the overall P_{flux} averages that contained a range of salinity gradients over the tested time period, although it could feasibly exist on a longer time interval.

Table 1.5: Mean P_{flux} from June 2010 to December 2012 (95% confidence limits) for the flux model calculated using the Texas rate-response curve and the rate-response curve reported by Tankersley et al. (1995) for megalopae collected in North Carolina at the Ship Channel Site at entrance to Mission-Aransas Estuary.

	Texas model	**Tankersley model**
Overall	-0.84(-1.22,-0.45)	-0.22(-0.45,-0.01)
Positive flux	4.11(3.63,4.59)	1.63(1.32,1.965)
Negative flux	-4.50(-4.89,-4.14)	-1.58(-1.92,-1.37)
Drought flux	-3.16(-3.83,-2.55)	-0.90(-1.20,-0.65)
Wet flux	2.75(1.39,4.05)	1.33(1.02,2.71)

Figure 1.8: Comparison of summarized ship channel model results with bootstrapped 95% confidence intervals between drought, normal years, and overall flux.



Models were also constructed for the Aransas Bay NERR site, which is within the estuary, to explore whether transport farther up estuary (and potentially to low salinity alternative nursery sites) is possible once megalopae have entered the estuary. At this site, the Texas model (1.20% day^{-1}, 95% confidence interval: 0.81, 4.59) for overall transport slightly outperforms the Tankersley model (0.76 % day^{-1}, 95% confidence interval: 0.38, 1.03), although most of the advantage lies again in efficient transport of Texas megalopae within wet periods (Table 1.6, Figure 1.9)

Table 1.6: Mean P_{flux} (% day^{-1}) from June 2010 to December 2012 (95% confidence limits) for the Aransas Bay flux model calculated using the Texas rate-response curve and the rate-response curve reported by Tankersley et al. (1995) for megalopae collected in North Carolina.

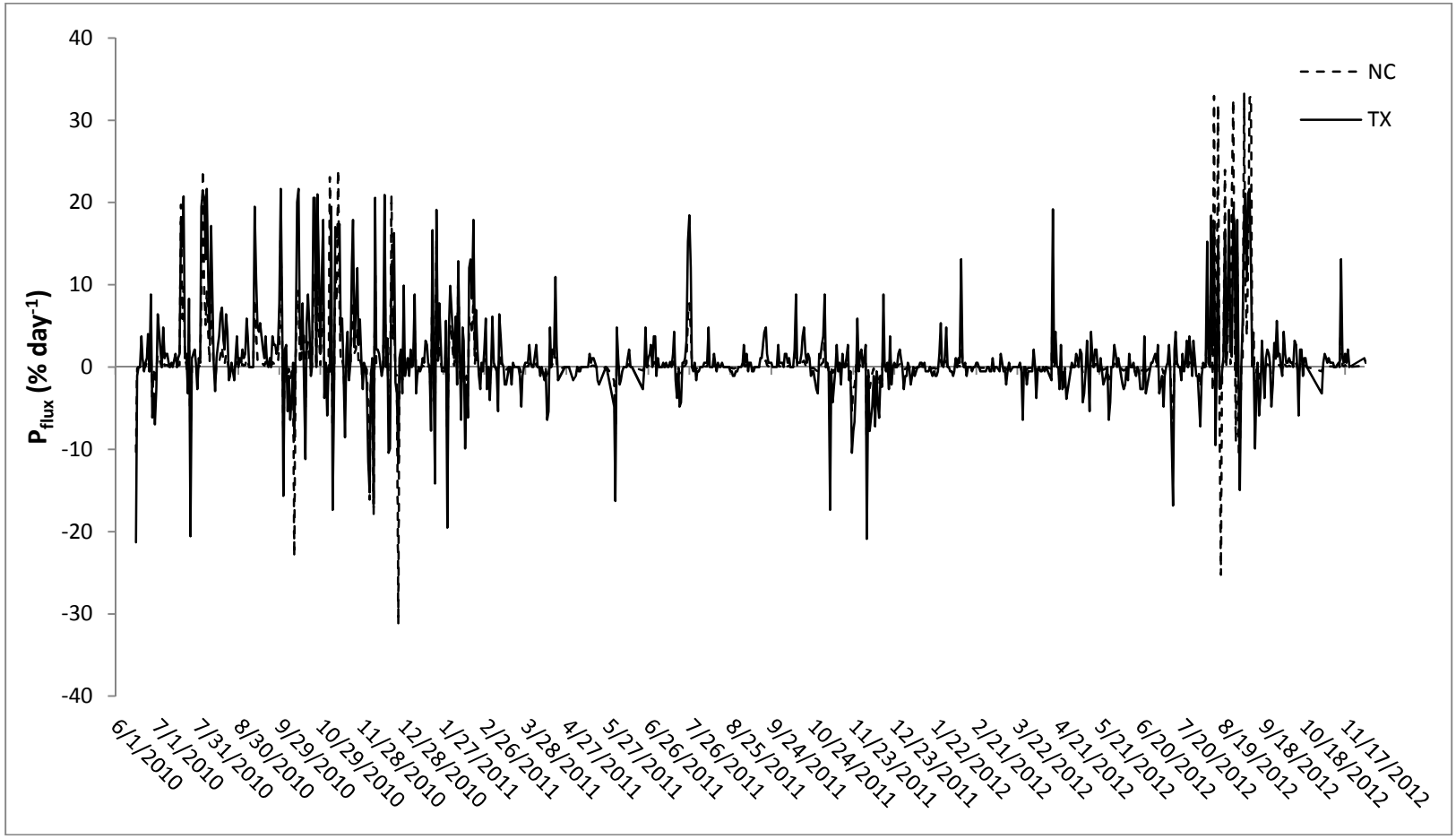
	Texas model	**Tankersley model**
Overall flux	1.20(0.81,4.59)	0.76(0.38,1.03)
Positive flux	4.40(3.88,4.96)	2.36(1.83,2.89)
Negative flux	-3.06(-3.59,-2.60)	-1.47(-1.97,-1.07)

The model outcomes at the Ship Channel site were relatively insensitive to the method that was used to calculate the rate of salinity change (Table 1.7). The direction of flux estimates were similar between salinity rate estimation methods, and the relative relationships between models (i.e. the Tankersley rate-response curve and the Texas rate-response curve) were consistent as well. Relative to the model based on moving average values of salinity change rates (used in the final model), estimates of P_{flux} were more extreme in the model based on 15 minute resolution rates, and were less extreme in the model based on hourly averages of rates. The smoothed model was retained, as smoothing minimized random noise while preserving underlying higher frequency signals.

Table 1.7: Mean P_{flux} (% day^{-1}) from June 2010 to December 2012 (95% confidence limits) for the flux model calculated using the Texas rate-response curve and the rate-response curve reported by Tankersley et al. (1995) for megalopae collected in North Carolina at the Ship Channel Site.

		Final model	**15 min. model**	**hourly model**
Tankersley model	Overall	-0.22(-0.45,-0.01)	-1.18(-2.08,-0.25)	-0.38(-0.73,-0.03)
	Positive flux	1.63(1.32,1.965)	9.26(8.62,10.35)	2.44(1.92,3.04)
	Negative flux	-1.58(-1.92,-1.37)	-10.6(-11.8,-9.53)	-2.51(-2.99,-2.09)
Texas model	Overall	-0.84(-1.22,-0.45)	-1.15(-1.81,-0.50)	-1.02(-1.49,-0.59)
	Positive flux	4.11(3.63,4.59)	8.58(7.90,9.29)	4.80(4.26,5.36)
	Negative flux	-4.50(-4.89,-4.14)	-9.46(-10.1,-8.87)	-5.39(-5.84,-4.94)

Figure 1.9: Results of behavioral transport model, predicting net flux as % per day, averaged by week from June 2010 to December 2012 past the within-estuary Aransas Bay site.



To investigate whether variations in the rates of salinity changes between nights play a role in blue crab megalpae recruitment in estuaries near where the Tankersley study was conducted, we applied the same model to data from the East Cribbing SWMP station in the North Carolina NERR, at the Cape Fear River Estuary. This estuary is 159 km from Beaufort, NC where the Tankersley study was originally conducted, and likely to experience similar climatic and tidally driven salinity conditions. Since this is a typical estuary that has reliable freshwater inflows, all increases in salinity were assumed to occur on the flood tide. Over the same 5 year time period, a net flux of (1.16 % day^{-1}) was reported for the Tankersley rate-response curve, with considerable variability. Surprisingly, the rate-response predicted for the TX megalopae was more reliable and would result in higher ingress due to a lower threshold for a response to a rate of salinity increase of Texas megalopae (2.16 % day^{-1}). These preliminary estimates do not take the seasonality of megalopae supply into account, which is known to vary in North Carolina, but is suggested to be year round in some Gulf Coast systems (Rabalais et al. 1995). Including seasonality in the flux estimates did not change the integrated outcomes considerably.

DISCUSSION

Experimental results

Our behavioral experiments have shown that megalopae collected from the entrance to the Mission-Aransas Estuary, Texas have a different response to various rates of salinity increase compared to megalopae in an experiment conducted in North Carolina (Tankersley et al. 1995, Figure 1.3). The responses of the Texas megalopae peaked at a

lower rate of salinity change and were more variable relative to the responses measured by Tankersley et al. (1995).

In the flood tide water experiment, both the magnitude of the response and the rate of salinity increase that elicited the maximum response varied widely among cohorts (Figure 1.3), but there was a significant peak response at a rate half that reported by Tankersley et al. (1995). This heightened sensitivity to lower rates of salinity increase may be adaptive to transport into the Mission-Aransas Estuary. Rates of salinity increase are generally lower at the entrance to the Mission-Aransas compared to estuaries in North Carolina with reliable freshwater inflows (CDMO).

In the comparison experiment, both Texas and North Carolina megalopae had a maximum positive response at the same rate reported in the original Tankersley et al. (1995) study (Figure 1.4). Although the rate that elicited the maximum response for Texas megalopae is higher in the comparison experiment than the average maximum positive response rate from the flood tide water experiment, the value falls well within the variance observed between cohorts. The North Carolina megalopae had a lower overall response than the megalopae collected in Texas, which may be a result of stress induced during shipping or the high salinity of the flood tide water used for acclimation prior to experimentation (37.8 ppt) compared to the salinity normally encountered by megalopae in North Carolina (32-35 ppt).

The variability of the behavioral response to salinity change reported in this study was much higher than the variability reported by Tankersley et al. (1995) despite using an equally powerful experimental design (Figure 1.3). Potential sources of variability lie in (1) experimental error and within-cohort behavior variability, (2) conditions of the water megalopae were tested in, and (3) intrinsic behaviors in megalopae that vary between cohorts. Experimental error and within-cohort variability are unlikely to contribute large

variability to our results, as the variance within treatments of the ocean salinity experiment (Figure 1.2) and in the comparison experiments (Figure 1.4) was generally low. Additionally, 30 megalopae were used in each treatment of each experiment, increasing the precision of proportion estimates to 3-4%. Megalopae within the same cohort presumably developed under the same environmental conditions, which contributes to the low variability within cohorts.

The potential variance introduced by differences in flood tide water properties (Table 1.3) collected on different dates was not tested as part of this study. There is some evidence that chemical cues associated with estuaries have a role in transport. Refractory chemical cues in estuarine water induce negative phototaxis in megalopae, which limits transport to the night (Forward and Rittschof 1994), and megalopae orient towards chemical cues associated with optimal habitats in flow (Forward et al. 2003) and on settlement substrates (Welch et al. 1997). Forward and Rittschof (1994) reported that megalopae are more active in offshore water compared to estuarine water, and this difference in kinesis could also function in transport, as demonstrated by Kingsford et al. (2002) in Japanese flounder. An increase in random movement will still result in some upward movement, as resting individuals on the benthos only have one vertical direction to move. Thus, an increase in random movement triggered by conditions during flood tide (high salinity for Japanese flounder, and potentially offshore seawater for megalopae), could result in some movement up into the water column, and transport on the flood tide. On the ebb tide, decreasing salinity for Japanese flounder and increasing estuarine cues for megalopae may result in decreased random movement and the increased likelihood of remaining in the boundary layer at the bottom, which would result in little horizontal movement during ebb tide but significant transport on the flood tide. Refractory humic substances and other dissolved organic carbon molecules that may

function as cues for megalopae tend to precipitate as salinity increases (Fox 1983), and the total DOC in the MANERR varies over drought and flood cycles, increasing with terrestrial carbon runoff during high freshwater inflow events, and decreasing during drought conditions (Bruesewitz et al., 2013). Differences in the chemistry of the flood tide water collected on different dates could plausibly have contributed to the variance noted in the flood tide experiment, but vary as a function of drought, salinity, mixing and other processes that cannot be adequately controlled in this study.

The results of the ocean salinity experiment isolated differences in intrinsic behaviors between cohorts as a source of variance, as water chemistry was controlled for, and within-cohort error was explicitly estimated using replicates (Figure 1.2). In the ocean water experiment, there were strong differences between cohorts, supporting the assumption that there are intrinsic differences in behavior that vary widely between cohorts. These "intrinsic" differences may have a genetic basis, or may be due to the influence of environmental conditions that modified their behavior before collection and testing as part of the experiment. The Mission-Aransas estuary lies at a major climatic gradient, as estuaries farther north and east along the Texas coast have stronger freshwater inflows and salinity gradients, while estuaries farther south tend to be hypersaline (e.g. Laguna Madre). The high variability measured here may be a result of offshore currents mixing populations of megalopae from different estuaries in variable proportions. As the salinity responses reported here are generally adaptive for transport into the Mission-Aransas via the Ship Channel, it is possible that megalopae from other estuaries also have behaviors ideal for transport into their home estuary. This variation may be a genetic selection event, or a result of phenotypic plasticity (e.g. megalopae are more likely to respond to rates of salinity change they have previously experienced during embryological development).

Model results

Flood and drought cycles

The model predicting P_{flux} is a useful tool for understanding the potential ecological implications and adaptive function of laboratory measured behaviors. The most notable model result is the strong role of flood and drought cycles on selective tidal stream transport behaviors (Figure 1.7), and potentially on recruitment success. While strong transport into the estuary was possible during wet or normal conditions, equally strong transport out of the estuary was predicted during drought conditions, showing that locally collected megalopae do not have salinity response behaviors that are adaptive for entry into the Mission-Aransas Estuary during drought (Figure 1.8). Some studies have noted an apparent link between blue crab populations and freshwater inflows (More 1969, Wilber 1993, Guillory 2000, Posey et al. 2005), but few have shown where the link lies. With the P_{flux} model, we have provided a plausible link for the long term correlations between freshwater inflows and blue crab abundance during selective tidal stream transport recruitment.

In the Aransas Ship Channel, megalopae have been caught in high abundances on the ebb tide, especially as a function of speed (Chapter 2), indicating that our model predicting export on the ebb tide is plausible. While most studies report megalopae in the plankton almost exclusively on nocturnal flood tides, transport of megalopae on the ebb tide has occasionally been reported. Paula et al (2004) reported significant transport of a variety of megalopae on the flood and ebb tide in a tropical mangrove system in Mozambique. Their system also had a shallow salinity gradient (< 5 ppt) and low rates of salinity changes ($-1x10^{-3}$ to $8.7x10^{-4}$ ppt s^{-1}), similar to the variably weak salinity gradients in the Mission-Aransas Estuary.

Even during drought conditions when our model indicates a net flux of megalopae out of the estuary (Figure 1.8), some flux of megalopae into the Mission-Aransas Estuary may still result from source-sink dynamics alone. Blue crab megalopae have a strong response to turbulence, and a large proportion of the population (~30%) will respond to increasing turbulence without any salinity cue (Welch and Forward 2001). Turbulent cues alone will not result in net flux into the estuary, as megalopae that are transported into the estuary on the flood tide will be carried back out again on the ebb tide. However, there are a variety of mechanisms (other than a salinity response in selective tidal stream transport) that may function to retain megalopae within estuaries after random transport.

Forward et al. (2003) showed that blue crab megalopae, especially megalopae that were close to metamorphosis into crab stages, can orient towards the scent of suitable settlement habitats such as seagrass beds. This "sink" function relies on the transport of megalopae close enough to settlement habitats for scent cues to be effective, and can still fail if by chance megalopae are not transported to these habitats. Megalopae are also less active in estuarine water (Forward and Rittschof 1994), and may be retained more in estuaries as a function of kinesis alone, and metamorphosis is accelerated in the presence of estuarine cues (Forward et al. 1997). If megalopae are more likely to settle within the estuary than outside of it, a megalopae entering the estuary is less likely to leave as it will have molted into a juvenile crab. Although net flux may continue in the absence of effective salinity cues, transport is certainly enhanced by strong salinity cues as our models show.

Comparison of transport within and into the estuary

A comparison of models of Texas megalopae transport (P_{flux}) showed that for Texas megalopae, transport farther up estuary at the Aransas Bay site was more reliable

than transport into the estuary through the Ship Channel (Figure 1.9, Table 1.6). This is likely due to less variable salinity signals within the estuary relative to the Ship Channel.

Although there is some evidence that salinity patterns in Aransas Bay are influenced by San Antonio Bay to the North, which generally has higher freshwater inflows, the salinity signals at the Ship Channel are more complex, as it exchanges between more bodies of water. These other bodies of water include the hypersaline Laguna Madre which has virtually no freshwater inflows, and the Corpus Christi Bay, whose freshwater inflows are highly reduced by two municipal reservoirs serving the freshwater needs of Corpus Christi, Texas. These often hypersaline bodies of water can contribute to the more dramatic increases in salinity on the ebb tide observed at the Ship Channel compared to Aransas Bay, which does not exchange as much water with Corpus Christi Bay and Laguna Madre (Kim 2009).

Complex salinity patterns are likely to be characteristic of other passes along the Texas coast. Inlets between the bays and the ocean are rare in the coastal bend region, with the nearest passes 90 km away to the North and 30 km to the South. Megalopae reaching the Mission-Aransas from other estuaries may have a single opportunity to reach an estuary before reaching the end of the megalopae stage. It is possible that inlet limitation plays a role in blue crab recruitment in the Mission Aransas Estuary, and that the complex salinity signals at inlets that receive flows from several bodies of water could further impede estuarine ingress of megalopae.

It is also possible that the salinity signals that fail to transport megalopae into the estuary may also fail to transport gravid female crabs out of the estuary to spawn, as females also undergo selective tidal stream transport (Darnell et al. 2009). Blue crab larvae require high salinities to complete development, and when high salinity conditions are present in the estuary, migration outside of the estuary may not be necessary. If

females do not leave the estuary to spawn, and the larvae are retained within the estuary, recruitment may not fail due to the weak salinity signals since the megalopae are already in the estuary. However, megalopae retained in the estuary may be subject to higher predation, and the occurrence of within-estuary spawns is uncertain. The overall impact of within-estuary spawns is currently unreported, and should be examined in future research.

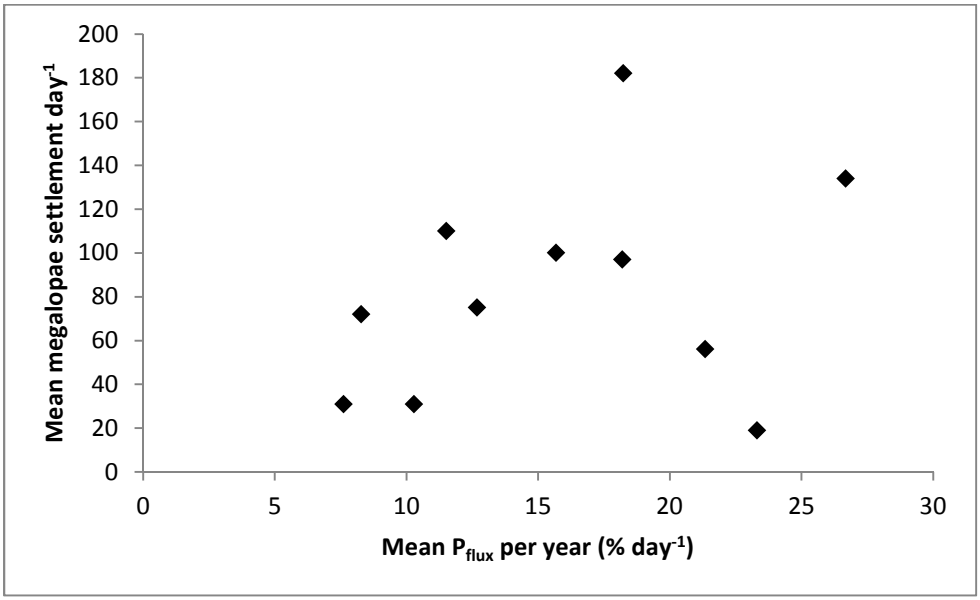
Comparison of Tankersley and Texas based models

The model fitted using the Tankersley rate-response curve predicted less overall export of megalopae from the estuary compared to the model created using the rate-response curve for Texas megalopae from this study. Further analysis of the models revealed that the difference in export was due to the overall weaker responses of Tankersley model megalopae to the rates of change experienced at the Ship Channel compared to the Texas megalopae (Figure 1.8). The difference in transport—either into or out of the estuary—shows that Texas megalopae are adapted to respond to the lower rates of salinity changes in Texas. However, the strong transport of Texas model megalopae out of the estuary during dry periods shows that the Texas megalopae salinity response is not drought adaptive. The Texas megalopae are just as likely to be transported out of the estuary during drought as they are to be transported into the estuary during wet periods. The model time frame encompasses one of the longest periods of drought in Texas, which has a clear impact on the ability of locally captured blue crabs to successfully recruit from the Gulf into the estuary.

To test the applicability of our model approach, we compared the results generated by the model using the Tankersley curve to blue crab megalopae settlement records. An 11 year long time series of blue crab megalopae settlement (widely regarded

to be correlated to the abundance of megalopae at the same site) is available for the Beaufort inlet and was analyzed to evaluate performance of our salinity-rate-response model as a recruitment indicator. While broad climatic patterns are likely to be similar in Beaufort and the Cape Fear River Estuary where the North Carolina NERR collects appropriate information for model formulation, the day to day fluctuations in salinity are probably not. The relationship between the predicted influx of megalopae using rates of salinity change and the actual average settlement was dome shaped (Figure 1.8). The dome shaped relationship suggests that when tidal salinity cues are weak, they may limit recruitment in this region, but when the tidal salinity cues are strong other processes (e.g. offshore availability) limit recruitment. Additionally, confounding variables such as turbulence may mask relationships between the supply of megalopae and settlement, as suggested by Amaral et al. (2008) and Bittler (Chapter 2).

Figure 1.10: Comparison of yearly aggregated salinity-behavior model results (P_{flux}, % transport day^{-1}) with yearly average settlement on hog's hair collectors for the Beaufort Inlet, North Carolina



Adaptive function of behaviors

The behavioral adaptations to salinity changes reported in this study are not unique to *Callinectes sapidus*. Salinity is a very important driver of selective transport in the meroplankton across a variety of taxa and geographic locations. In the green shore crab, *Carcinus maenas*, circatidal rhythms can be entrained with abrupt changes in salinity to enable selective tidal stream transport (Taylor and Naylor 1977), and accordingly, *C. maenas* megalopae are most abundant in high flood tide salinities in the field. Japanese flounder are more active in higher salinity waters, which facilitates transport on the flood tide (Kingsford et al. 2002), and models have suggested that salinity cued STST results in more rapid and effective transport of brown shrimp (*Crangon crangon*) into estuaries in the North Sea (Daewel et al 2011). Indeed, other modeling studies that were complemented with behavioral observations have shown that disparate behaviors can have huge consequences for dispersal. North et al. (2008)

compared a simple behavior of oyster larvae encountering a halocline in two species: *Crassostrea virginica* and *C. ariakensis*. C. *virginica* swam up in response to a halocline, and *C. ariakensis* swam down in preliminary behavioral experiments. The simple difference in behavior resulted in huge differences in dispersal, and was more influential in the dispersal model than hydrodynamic flows.

The geographically disparate salinity rate-response behaviors observed in this study within a single species appear to be adaptive for transport under geographically disparate estuarine conditions. There is some genetic structuring of blue crab populations that could feasibly support such local adaptation. While this structuring is significant, it does not appear to follow any geographic trend (Kordos and Burton 1993, McMillen-Jackson 2004). In a study comparing population genetics of megalopae and adult blue crabs along the Texas coast, Kordos and Burton (1993) found that the genetic signature of megalopae did not always match the adult blue crabs found in an area. They also found that while megalopae tended to have genetic structuring in the summer months, in winter the population was well mixed. The temporal variability in megalopae genetics may be an underlying mechanism supporting the wide variance we observed in our rate response curves measured in cohorts separated by time. The authors suggest that gene flow between populations is lower than the forces of random drift or natural selection, resulting in the genetic structuring observed. If natural selection is one of the structuring forces in blue crab populations, selection based on salinity-driven recruitment behaviors may allow for some local adaptation. Additionally, if populations are somewhat isolated by megalopae retention within an estuary, this local adaptation process can be accelerated. There is some evidence that most successfully recruiting megalopae are retained within a single system in the Mississippi Bight (Johnson and Perry 1999) and the Chesapeake Bay (Roman and Bolcourt 1999) using a hydrodynamic modeling approach.

However, this retention is at odds with the genetic mismatch reported between megalopae and adult blue crabs in Texas (Kordos and Burton 1993), and is an unresolved issue.

Local adaptation in marine and estuarine species is not unprecedented. Colin and Hans (2002) reported geographic gradients in the copepod *Acartia tonsa*'s tolerance to toxins of the dinoflagellate *Alexandrium*. While *A. tonsa* collected from estuaries where *Alexandrium* is present could survive the toxins in assays, naïve *A. tonsa* collected from estuaries outside of *Alexandrium* species ranges were much more sensitive. The authors postulate that co-evolution of *A. tonsa* and toxic dinoflagellates within systems drove the adaptation to the toxins over time, and that this process was relatively rapid due to the short generation times of *A. tonsa*, and potentially low gene flow between estuaries of this strictly estuarine copepod species. However, locally adaptive behaviors are rarely studied in the marine environment, and the drivers of these behaviors are generally unknown.

Implications for drought cycles and freshwater management

This study has provided evidence that net transport of blue crab megalopae out of the estuary on the ebb tide during periods of drought could be relatively substantial due to the rates of salinity change the megalopae experience on outgoing tides when salinities in the estuary are high (Figure 1.7, Table 1.5). Within a given estuary, rates of salinity change can be related to salinity gradients over the long term (Figure 1.5). These salinity gradients are driven by the balance of freshwater inputs with evaporation and mixing processes, and are linked to drought cycles and human alterations of freshwater inflows.

When freshwater inflows are lowered to a point when the appropriate rates of salinity changes are no longer reached to elicit a vertical response from megalopae during selective tidal stream transport, recruitment of blue crab larvae into the estuary will be

impaired, as will movement farther up estuary to suitable nursery sites. Lower blue crab recruitment success to favorable nursery grounds may lead to declines in the population within the estuary. While it is possible that adult females remain in the estuary to spawn under high salinity conditions, the understanding of this process is incomplete. The established importance of selective tidal stream transport in recruitment of blue crabs and the results of this study lend support to reports of a link between blue crab populations and freshwater inflows reported in the literature.

Chapter 2: To Settle or Not to Settle? The Efficiency of Hog's Hair Collectors Under Varying Flow Conditions

ABSTRACT

Hog's hair collectors are simple artificial settlement substrates widely used to quantify brachyuran megalopae settlement. Despite the wide use of hog's hair collectors, few studies have attempted to quantify settlement in terms of megalopae transport or abundance. The studies that have compared settlement to planktonic abundance of megalopae generally have reported poor correlations, possibly due to different temporal collection scales. The results of a laboratory flume and field settlement study showed that settlement is generally highest at low current velocities, but that settlement is still possible even at high current velocities. The application of tomographic particle image velocimetry (PIV) quantified the small scale flow patterns behind collectors, and showed that an area of lowered turbulent kinetic energy (TKE) is maintained even as current speeds increase (up to 38 cm s^{-1}, the maximum velocity measured), and that this zone of lowered TKE may be a critical signal stimulating megalopae to settle. The application of an encounter rate model explained the processes driving lower settlement as a decreasing ability of megalopae to sense the collector as current velocity and turbulence increase, a phenomena observed in planktonic predator-prey interactions. Simple and effective means of accurately interpreting hog's hair collector data as either transport or as an index of megalopae abundance are presented.

INTRODUCTION

Hog's hair collectors are passive artificial settlement substrates that are often constructed of air conditioning filters wrapped around a section of PVC pipe (Metcalf et al. 1995). These practical and inexpensive samplers have been widely used to quantify

settlement of postlarval brachyuran crabs (megalopae), including the Blue Crab *Callinectes sapidus*. Hog's hair collectors are used on a variety of spatial scales (Olmi 1990, Flores et al. 2002), to capture highly episodic recruitment events (Goodrich et al. 1989, Rabalais et al. 1995) and are regarded as being well correlated to the actual planktonic supply of megalopae (Lipcius et al. 1990, Ogburn et al. 2009). Despite the wide use of these samplers, few studies have critically examined their limitations.

Some studies have found poor correlations between the planktonic abundance of megalopae and the number collected on hog's hair collectors (Lipcius et al. 1990, Olmi et al. 1990, Moksnes and Wennhage 2001). For example, Moksnes and Wennhage (2001) found no correlation between the settlement of shore crabs on artificial substrates and their abundance in plankton tows in a Swedish estuary. They attributed the lack of correlation to the fact that artificial settlement substrates integrate the abundance of megalopae over the entire duration the collectors are deployed, while plankton tows represent a mere snapshot that does not capture the variation in abundance that takes place over the entire night. Amaral et al. (2007) explicitly addressed these issues by measuring both the planktonic abundance and settlement of *Carcinas maenas* megalopae in a Portugal estuary at high temporal resolution. They found that settlement on bottom-deployed settlement substrates was decoupled from the supply of megalopae measured using plankton nets, suggesting that turbulence may decouple settlement from megalopae supply. While several studies have attempted to correlate settlement with abundance and flux, none have estimated planktonic abundance and settlement at precisely the same time frame and spatial scales. With instantaneous measures of settlement and abundance, precise estimates of efficiency can be determined.

While megalopae counts taken from hog's hair collectors deployed for 24 hours are widely regarded as integrating the supply of megalopae over the course of a day, Mocksnes and Wennhage (2001) reported losses of up to 83% of settling shore crabs from artificial substrates deployed for more than 12 hours. Tankersley et al. (2002) sampled hog's hair collectors hourly throughout the night in order to identify when peak settlement occurred in *Callinectes sapidus*. When compared to the number of megalopae collected over the course of the night from hourly deployments, significantly fewer megalopae accumulated on a collector left unsampled the whole night. They attributed this discrepancy to predation on the collectors, or to behavioral responses that trigger swimming downward in some megalopae at sunrise.

Another element that may confound interpretations of megalopal abundance measured from hog's hair collectors is behavioral selection for substrates that may vary depending on molt stage. Several studies have noted differences in megalopal stage and time to metamorphosis between megalopae collected in the plankton or from collectors at the same site (Lipcius et al. 1990, Metcalf and Lipcius 1992), although this trend is not universal (Olmi et al. 1990, Hasek and Rabalais 2001). All megalopae are thigmotactic (i.e. attracted to surfaces such as the hog's hair collectors), but the differences between megalopae collected from hog's hair collectors and from the plankton raise questions of whether thigmotactic settlement behavior is uniform among all stages of the megalopae. It is also possible that settlement itself is a potential stimulus that can decrease the time to metamorphosis in blue crabs, potentially explaining the observed differences, although Forward et al. (1996) found that structural cues alone did not shorten the time to metamorphosis. It is still unresolved whether these differences are due to the stronger behavioral drive to settle in later stages of megalopae that are about to metamorphose, or

if the act of settling on a substrate itself cues the megalopae to move into the later megalopal stages and shorten the time to metamorphosis (Metcalf and Lipcius 1992, Hasek and Rabalais 2001).

Other variables in the environment may physically remove megalopae from collectors or be behavioral cues that cause megalopae to emigrate from collectors. However, we could find no studies that quantify the effects of current velocity or turbulence on retention of megalopae on collectors, although some studies have found correlations with broad environmental conditions (De Vries et al. 1994, Olmi 1995, Tankersley et al. 2002, Forward et al. 2004). There is likely a threshold velocity or turbulence intensity above which megalopae are physically unable to settle onto the collectors. However, it is unknown what these conditions are, and whether they frequently occur in nature. Additionally, megalopae are known to have swimming responses to turbulence (Welch and Forward 2001) and changes in salinity (Tankersley et al. 1995) which vary over the course of tidal cycles that would occur during collector deployment. If settlement is not a permanent process, and megalopae and young juveniles continue to move between habitats in the plankton (as Etherington and Eggleston 2000 suggest), then settlers are likely to emigrate from the collectors in response to changing environmental conditions. In this case, changing environmental conditions are likely to confound interpretations of megalopal abundance as measured using settlement substrates.

The effects of current speed and turbulence on megalopae settlement can be considered with an encounter rate model. As turbulence or current speed increases, one would expect for the encounter rate to increase linearly, and for more megalopae to settle on the collectors as a function of increased delivery. However, at higher levels of current

speed and turbulence, the retention of megalopae on the collectors may be expected to break down as megalopae are unable to settle or hold onto the substrate (or as they choose to move, as described previously). The interaction between increasing delivery with increasing current speed and increasing loss past some speed and turbulence threshold predicts a dome shaped relationship between turbulence and settlement. This relationship has been successfully applied to the probability of larval fish feeding across a scale of turbulence, where the encounter rate increases with turbulence, but the pursuit success decreases (MacKenzie et al. 1994). In this case, the collector can be treated like the "predator", but it is the ability of the megalopae "prey" to find and settle on the collector that determines how efficiency changes with turbulence.

No studies to date have examined how the efficiency of hog's hair collectors changes with turbulence, characterized the small scale flow environment around the collectors, or observed how megalopae interact with the collectors. This study will investigate how megalopae interact with hog's hair collectors using a model species, the Blue Crab *Callinectes sapidus*, and will resolve some of the issues posed for using artificial settlement substrates to quantify abundance and settlement. We will examine the influence of current velocity on artificial substrate settlement using an encounter rate model, and will determine the sampling efficiency in both field and laboratory experiments. Additionally, the influences of small scale flows behind the collectors on behavioral interactions will be examined across a range of current speeds. Our hypotheses are:

(1) The efficiency of collectors is comparable to the encounter rate, and that every megalopae that passively encounters the collectors will settle

(2) The efficiency of the collectors does not change as current speed or turbulence changes

(3) The flow characteristics behind the collector do not change with current speed, and that megalopae do not approach collectors from downstream using these signals.

METHODS

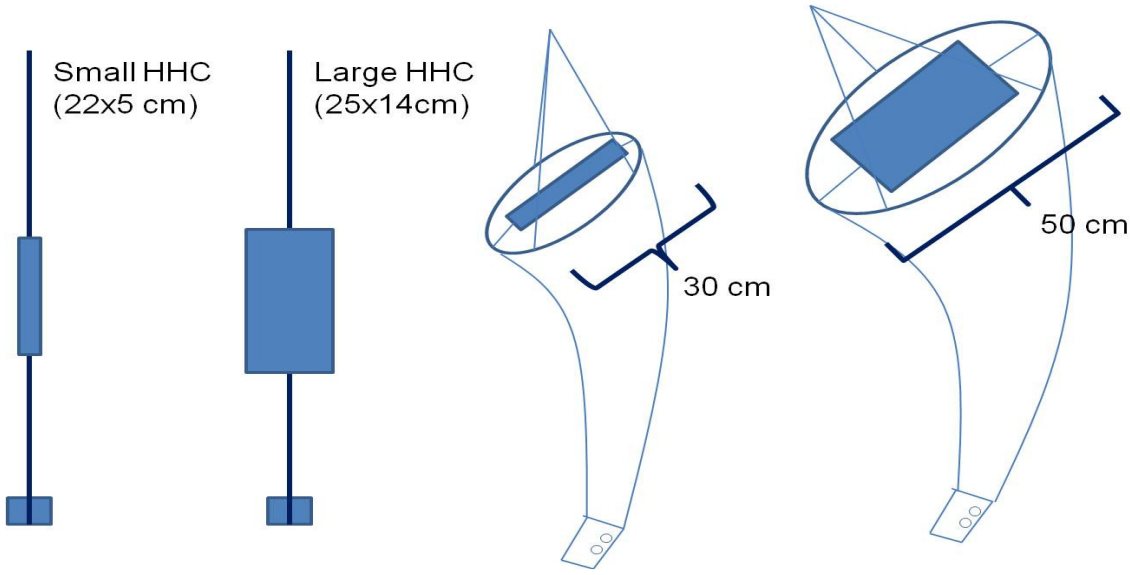
Field experiment

The efficiency of hog's hair collectors was tested in the field by sampling collectors every 1-2 hours throughout the night on the incoming tide, when megalopae are most abundant (Mense and Wenner 1989), at the University of Texas Marine Science Institute (UTMSI) ship channel pier. The Port Aransas ship channel experiences strong tidally-driven currents with velocities that range from 0 to 1.5 m s^{-1} (Scheef, unpublished data, Min, unpublished data). Multiple samples were taken each night to capture the effects of variable current speeds while controlling for factors that may vary between sampling nights (e.g. offshore availability, salinity gradients, etc.). Sampling was conducted on 10 nights, every 1-2 months from September 2012 to October 2013.

Hog's hair collectors were constructed of PVC pipe wrapped with air conditioning filter media secured with rubber bands. Hog's hair collectors were fastened within the mouth of plankton nets, such that all megalopae that did not settle while passing by the collectors would be sampled and estimates of efficiency could be generated. While the collectors likely impede flow into the plankton net, the relative ratio of settling megalopae to non-settling megalopae is unlikely to be changed by this sampling artifact. Two sizes of collectors and nets were used: a large (25 cm long x 14

cm diameter) collector fastened in a 0.5 m diameter, 500 µm mesh plankton net (0.5 m diameter, 330 µm mesh plankton net before October 2012); and a scaled down collector (22 cm long x 5 cm diameter) fastened in a 30 cm diameter, 440 µm mesh plankton net (Figure 2.1). The smaller collector was tested in the field to evaluate whether collector size influenced estimates of settlement in flume experiments (see below). Additionally, both large and small collectors were suspended near the surface without a plankton net to account for the possibility of artifacts of the plankton net on settlement. Sampling was generally only conducted when the current was strong enough to suspend the nets in the current, but the entire flood tide, including slack tide, was sampled on October 18, 2013. At slack tide, only the large and small collectors without nets were used, and a plankton net (30 cm diameter, 330 µm mesh) was towed up and down the pier for the duration of sampling to attain an estimate of the planktonic abundance of megalopae.

Figure 2.1: Sampling apparatus for field experiments, including two hog's hair collectors suspended on rope in near water surface, and two collectors secured in the mouth of plankton nets towed near the surface.



The entire sampling apparatus (Figure 2.1) was deployed for 15 minutes from September to November 2012, and the sampling time was increased to 30 minutes after January 2013 to increase the low sampling size of settling megalopae. After each deployment, collectors were rinsed with freshwater and visually inspected for settled megalopae. Plankton samples were first strained through a 5 mm mesh to remove large debris, then concentrated on a 500 µm mesh sieve in the field. Megalopae are generally 2-3 mm in size, and pass readily through the coarse mesh but not through the finer sieve. Plankton samples and all megalopae removed from collectors were preserved in 50% ethanol solution and counted in the laboratory. In cases where extremely high numbers of megalopae were present in the plankton samples, a subset of at least 200 individuals was counted and extrapolated to estimate the densities in the whole sample following standard zooplankton analysis procedures. Plankton tow samples were subsampled with a plankton splitter (Wildco Folsom Plankton Splitter).

Current velocity was measured by OkeanoLog Seahorse tilt current secured about 1 m below the surface on a tide trap throughout sampling. The abundance of megalopae in the plankton was estimated by dividing the raw counts by the current velocity, deployment time and entrance surface areas of the plankton net. Raw counts of megalopae were compared between plankton nets with and without collectors (Figure 2.1) to ensure estimates of megalopae abundance were reasonable.

The relative abundance of megalopae in the plankton, the number of settling megalopae, and the settlement rate (the number settled divided by the number of megalopae available in the plankton) were compared at various current speeds during the flood tide. Data was analyzed using an ANCOVA type model, treating sample night as a fixed factor and current velocity as a covariate.

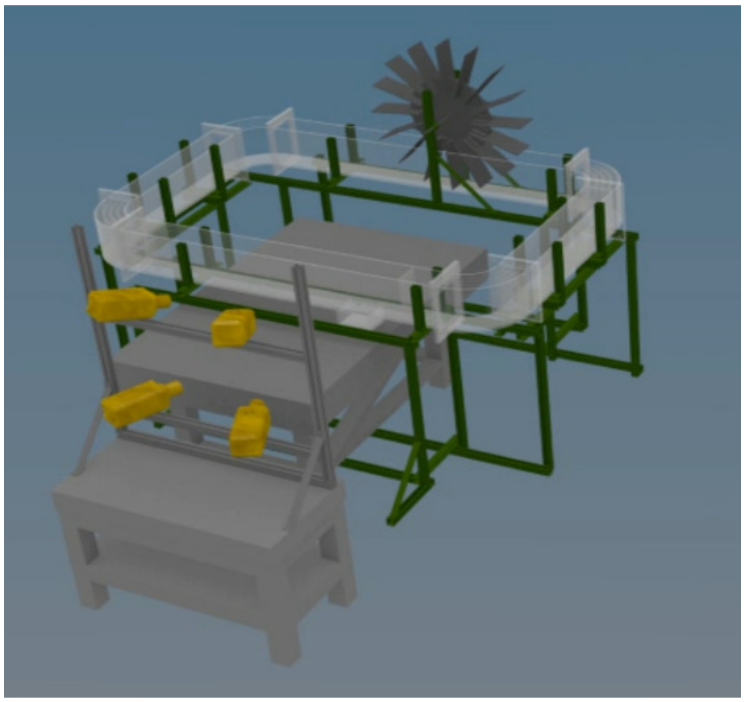
Flume experiment

The effects of current velocity on megalopae settlement were observed and tested in a flume experiment. Megalopae were captured on the flood tide from 5-6:00 am on September 14, 2012 from the University of Texas Research Pier in the Port Aransas Ship Channel (27º 50' N, 97º 3' W) with two 0.5 m diameter 153 µm mesh plankton nets. Megalopae were held in seawater until experiments began. Experiments were conducted under light conditions from 3-6:00 pm, and under dark conditions from 8-11:00 pm. Both experiments were conducted during ebb tide in the field, during a diurnal tidal cycle. Tides in the Western Gulf of Mexico are mixed between semidiurnal and diurnal phases and vary week to week.

Fifty megalopae were allowed to circulate in a 265 L flume (Figure 2.2) and adjust to experimental conditions for 10 minutes before a scaled-down hog's hair collector (5 cm diameter) was added to the experimental section of the flume. A 500 µm mesh was simultaneously added just downstream to capture megalopae that did not settle on the collector, so each megalopae only had one chance to settle. The collector was recorded by a Cohu 6500 camera with a Sony Digital 8 Video Walkman for 5 minutes, giving each megalopae ample opportunity to contact the collector (3.6 minutes to circulate the entire flume at the lowest current speed). The collector was rinsed with freshwater onto a 500 µm mesh sieve, and visually inspected for settling megalopae, then the flume speed was increased to 38 cm s^{-1} to flush any remaining megalopae onto the mesh. The experimental series evaluated a range of velocities: 3.5 cm s^{-1}, 20 cm s^{-1} and 38.3 cm s^{-1} under fluorescent light and in darkness illuminated by infrared LEDs (>850 nm), with three replicates per treatment. Estimates of settling rate were calculated by dividing the number of settling megalopae by the sum of settling megalopae and the

number of megalopae re-collected on the mesh at the end of each trial after flushing flume for 3 minutes (at 38.5 cm s^{-1}). Data were analyzed using linear regression so estimates could be extrapolated to intermediate current speeds.

Figure 2.2: Flume used in settlement experiment and for tomographic PIV characterization of small scale flows behind collectors (Image by D. Adhikari).



Tomographic PIV characterization of small scale flows around collector

The small scale flow characteristics just behind the hog's hair collectors were quantified using Tomographic Particle Image Velocimetry (PIV) across three current speeds (3.5 cm s^{-1}, 17.4 cm s^{-1}, 31.3 cm s^{-1}). The current speeds measured were similar, but not identical to the speeds tested in the flume experiment. A paddle wheel circulated seawater around a 75 gallon capacity flume with a channel width of 150 mm filled to a depth of 150 mm (Figure 2.2). A honeycomb 1.3 meters upstream reduced turbulence

created by the paddle and bends in the channel before perturbation by a 50 mm diameter scaled down hog's hair collector (the same collector used in flume settlement experiments). The water was seeded with 55 µm diameter titanium dioxide filled polyamide particles (200 particles cm^{-3}) used to track water movements in the 2x8x4 cm observation volume immediately behind the collector. Water movements were recorded at 66 frames per second (current speed 3.5 cm s^{-1}), 285 frames per second (current speed 17.4 cm s^{-1}) and 600 frames per second (current speed 31.3 cm s^{-1}) by four high speed cameras (1280 x 800 pixels, Phantom v. 210, Vision Research Inc.), illuminated by an Oxford firefly laser (808 nm; 300 W). Different frame rates were used for each current speed to ensure that each flow was recorded for long enough to characterize at least 10 eddy shedding events (as calculated by an ideal cylinder). The videos were analyzed with the MLOS-SMART algorithm in DaVis 8.0, and data visualized in TechPlot. For further details see Adhikari and Longmire (2013).

Encounter rate model

The encounter rate model is a useful framework for comparing velocity-dependent settlement processes when direct estimates of the proportion settling are unavailable (such as at slack tide). If we assume that megalopae are randomly distributed dimensionless points that move randomly, Gerritsen and Strickler (1977) present a formula for estimating the "contact rate" as the number of prey (the megalopae) contacting a dimensionless predator (the collector):

$$Contact\ rate = \pi * R^2 * N * \frac{v_{prey}^2 + 3v_{pred}^2}{3v_{pred}}$$

Where R is the search radius of megalopae contacting the collector, N is the abundance of megalopae, v_{pred} is the speed of the collector (in this case, the speed of the current since the collector is held at a fixed point), and v_{prey} is the velocity of the megalopae, set to 10 cm s^{-1} (based on the maximum sustained swimming speed measured by Luckenback and Orth 1992). This formula holds for conditions when $v_{pred}>v_{prey}$, in this case, when current speeds are greater than 10 cm s^{-1}, which encompasses our entire sampling range (even at slack tide).

In our calculation, we assume that R is unknown, and that every megalopae contacting the collector settles. As such, we set contact rate equal to settlement divided by the length of deployment and solve for R.

RESULTS

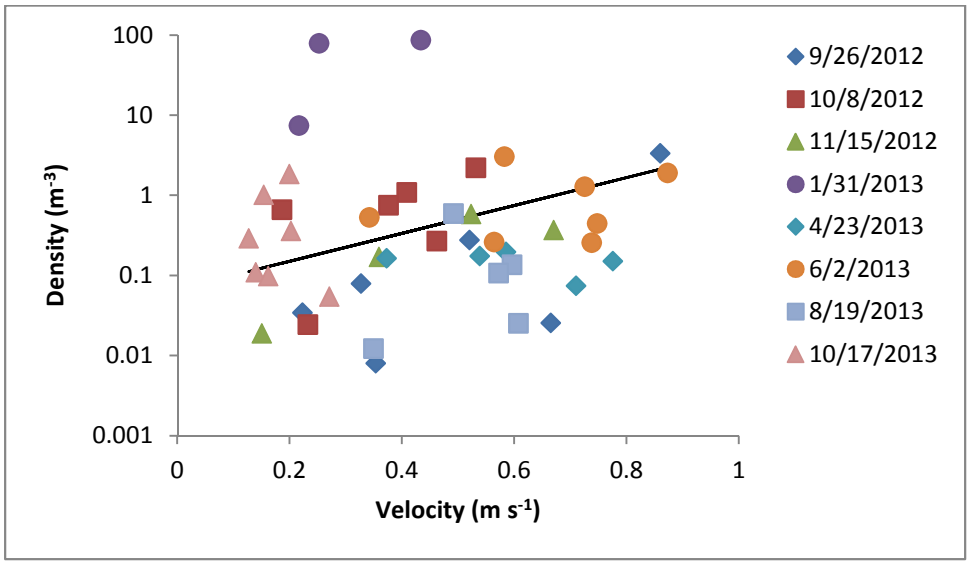
Field sampling

A one way Kruskal Wallis test determined that there were no differences in settlement based on the size of collector (large or small) or the presence of a plankton net ($p=0.1742$, $df=3$). The test did not find significant differences between any groups, indicating that total settlement on collectors suspended in nets were reasonable, and that artifacts of collector size were unimportant in the context of this study. As an additional precaution, the proportion of megalopae settling on the small collector in the plankton net and the large collector in the plankton net were also compared to detect artifacts of size, these results did not show a significant difference (paired Wilcox test of means, $p=0.6724$). Thus, settlement and plankton estimates from each sized collector suspended in the plankton nets were pooled for the remaining analyses.

The correlation between the plankton abundance in the nets with and without collectors was strong, with linear regressions predicting an increase of 0.967%

megalopae in the plankton net with the collector for a one percent increase in abundance in the net with no collector ($p<0.001$). The slope of this relationship is very close to the expected 1:1 relationship, indicating that estimates of plankton abundance from the net with a collector suspended in the mouth are accurate. Thus, plankton densities from the plankton nets with collectors were used for the rest of the analysis.

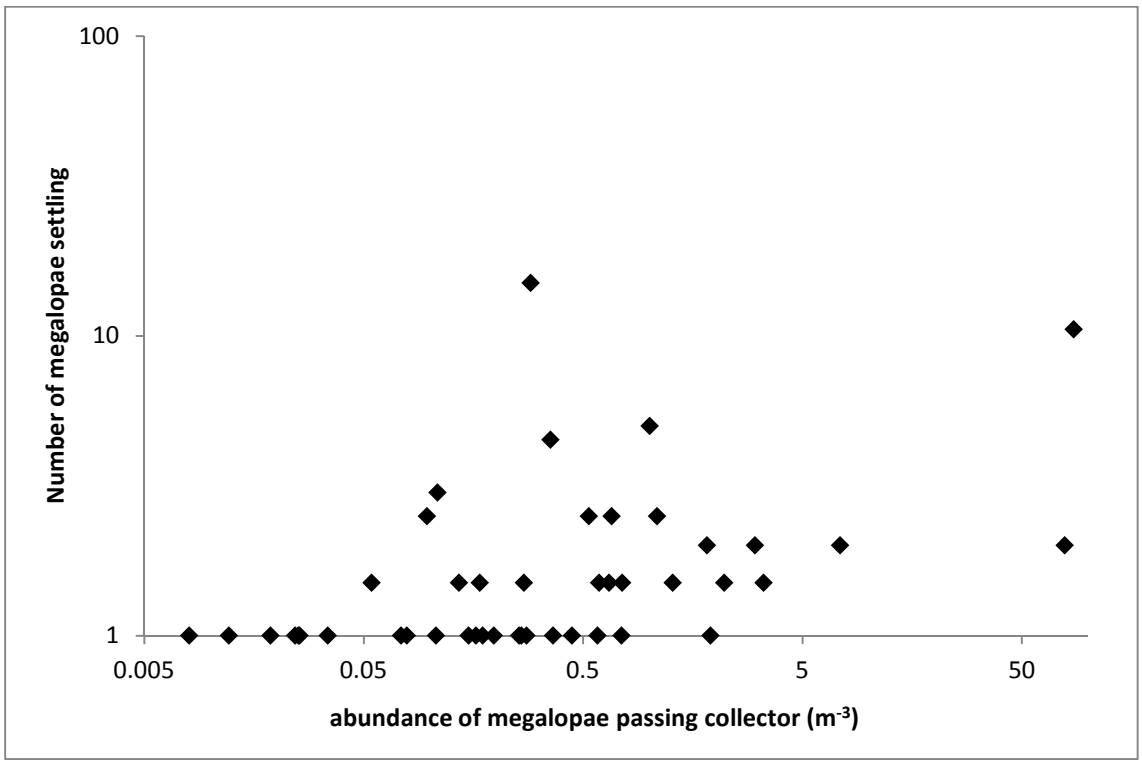
Figure 2.3: Abundance of *C. sapidus* megalopae (m^{-3}) by current velocity ($m\ s^{-1}$) on all sampling nights. Trendline shows the relationship estimated by a global regression ($p<0.001$).



The abundance of megalopae in the plankton (m^{-3}) increased by 212% per one meter per second increase in current velocity ($p=0.001$, Figure 2.3). The relationship between megalopae abundance and current velocity was consistent between nights, as indicated by the non-significant interaction term ($p=0.456$). There were significant differences in overall abundance of megalopae on different sampling nights ($p<0.001$), with model intercepts predicting densities at slack tide varying between 10.6 megalopae m^{-3} in January 2013 and $8.8x10^{-3}$ megalopae m^{-3} in August 2013. No temporal pattern of

megalopae abundance was evident, except for an extreme peak in winter (January 2013) relative to the lower abundances through the rest of the year (Figure 2.3).

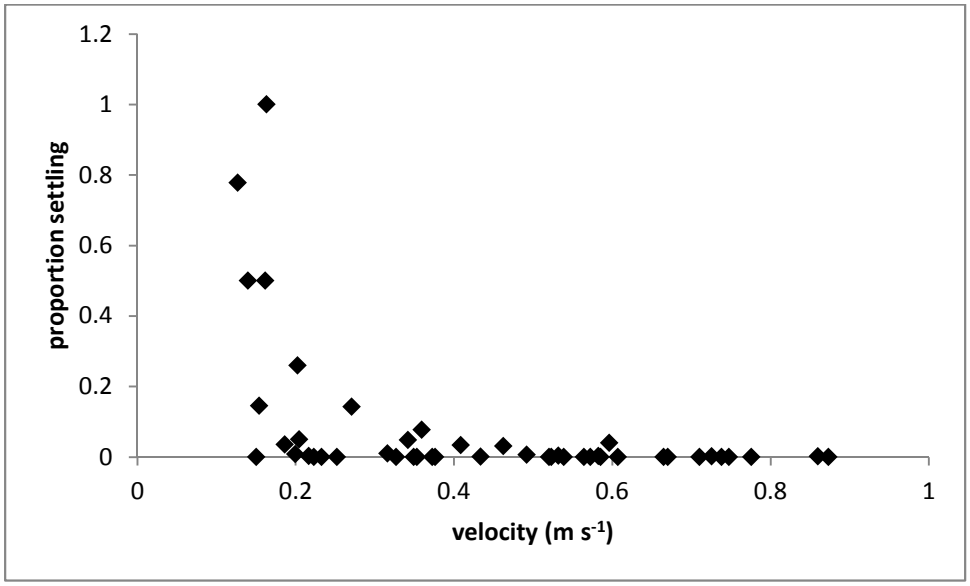
Figure 2.4: Settlement (total number of megalopae settling) versus the abundance of megalopae in the plankton (individuals per m^3). Settlement counts are (1+Settlement) to plot zero values on a log axis.



There was a general positive relationship between the total number of megalopae settling on collectors and abundance of megalopae in the plankton, although the relationship appears to be more bell-shaped at abundances below 5 megalopae m^{-3}. The number of settlers and abundance estimates were highly skewed, but natural log transformations were successful in establishing normality in the residuals of a linear model predicting total settlement from the number of passing megalopae (Figure 2.4). An increase of 0.16 % in settlement is expected for every 1% increase in megalopae

abundance, controlling for the effects of current velocity ($p<0.001$), but a decrease of 1.08 % settlement is expected for a 1 m s^{-1} increase in current velocity. Between these two variables, 48.4% of the variance in settlement was explained. There were significant differences in total settlement between sampling nights ($p<0.001$), but the relationship between settlement, abundance and velocity was consistent between nights, as validated by the non-significant interaction term.

Figure 2.5: Proportion of megalopae settling compared to current speed (m s^{-1}). Proportions calculated based on the number of settlers compared to the number of megalopae collected in the adjacent plankton net.



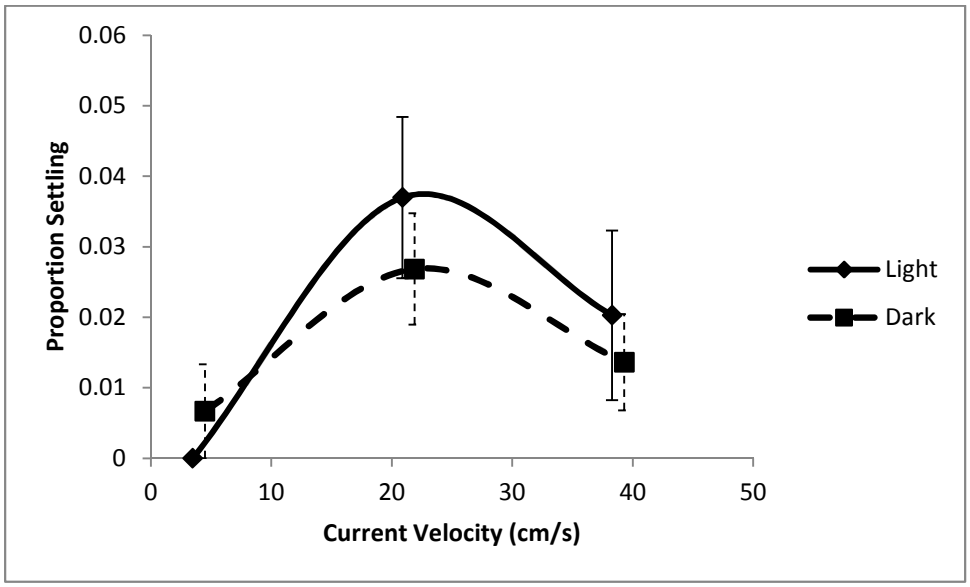
The relationship between proportion of megalopae settling and current speed appears to exponentially increase to a maximum value of one as the current speed decreases, and is significantly explained by a logistic regression model ($p<0.001$, Figure 2.5). Settlement at current speeds above 0.2 m s^{-1} tended to be below the contact rate from random chance alone (20%, based on the area of the plankton net occupied by the collector), while settlement at current speeds below 0.2 m s^{-1} generally surpassed the expected encounter rate.

On October 18, 2013 we sampled for the entire flood tide cycle, including slack tide. The study design using plankton nets attached to collectors is ineffective when the tide is too weak to suspend the net in the current. At slack tide we could not directly estimate the proportion of megalopae settling, and instead estimated plankton abundance and settlement on collectors independently. The estimates of abundance at slack tide were 0.15±0.04 megalopae m^{-3}, which was close to the estimate of the abundance of megalopae at a current speed of zero from the intercept of the regression between plankton abundance and velocity (0.143 megalopae m^{-3})

Flume experiment

The relationship between the proportion of megalopae settling and current velocity was re-examined in an experimental flume. Under the controlled experimental conditions, a significant dome-shaped relationship was observed that was explained well by a quadratic regression ($p=0.02$), predicting maximal settlement at current speeds between 13 and 43 cm s^{-1}. There was no difference in the nature of this relationship under fluorescent light or dark conditions (Figure 2.6).

Figure 2.6: Proportion of megalopae settling in dark and under floursecent light conditions in a 75 gallon flume experiment.



In addition to the settlement-velocity curve obtained in the flume experiments, the behavior of the megalopae interacting with the collectors was observed in the X-Y plane. Megalopae actively approached the collectors from both upstream (41% of approaches) and downstream (59% of approaches), with no significant variation in the proportion of approaches from up or downstream with current velocity ($p=0.653$). The proportion of megalopae that interacted with the collector varied significantly with current velocity ($p=0.0127$), with low interaction at the highest current velocities (7.6%) and the highest interactions at the lowest current velocity (12.6%). Megalopae generally slowed down or maintained position when near the collector, sometimes swimming towards and physically contacting the collector when interacting with it (representative paths shown in Figure 2.7).

Figure 2.7: Paths of megalopae in the X-Y plane under low (3.5 cm s^{-1}, A), medium (20 cm s^{-1}, B) and high (38 cm s^{-1}, C) current speeds observed in dark flume conditions. Collector position is shown in gray, and water flows in the negative x-direction (from 14 towards 0).

Figure 2.7A: Low current speed (3.5 cm s^{-1})

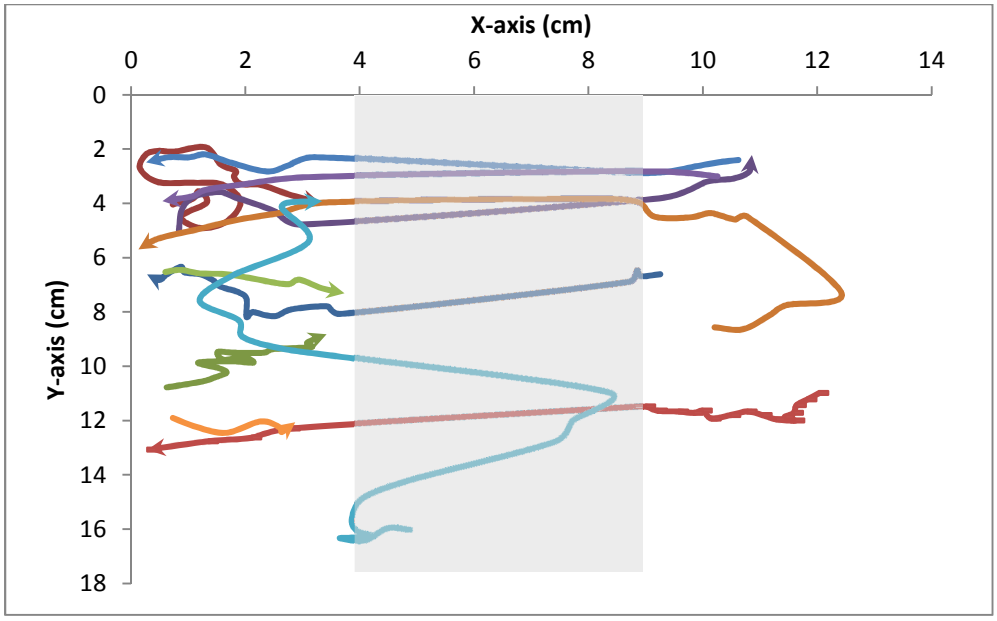


Figure 2.7B: Medium current speed (17.4 cm^{-1})

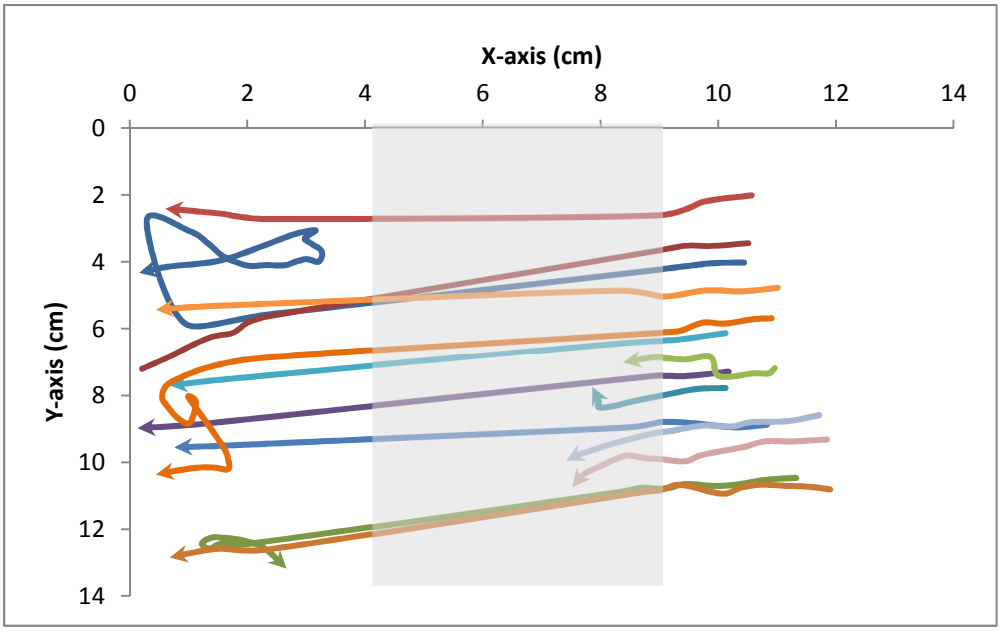
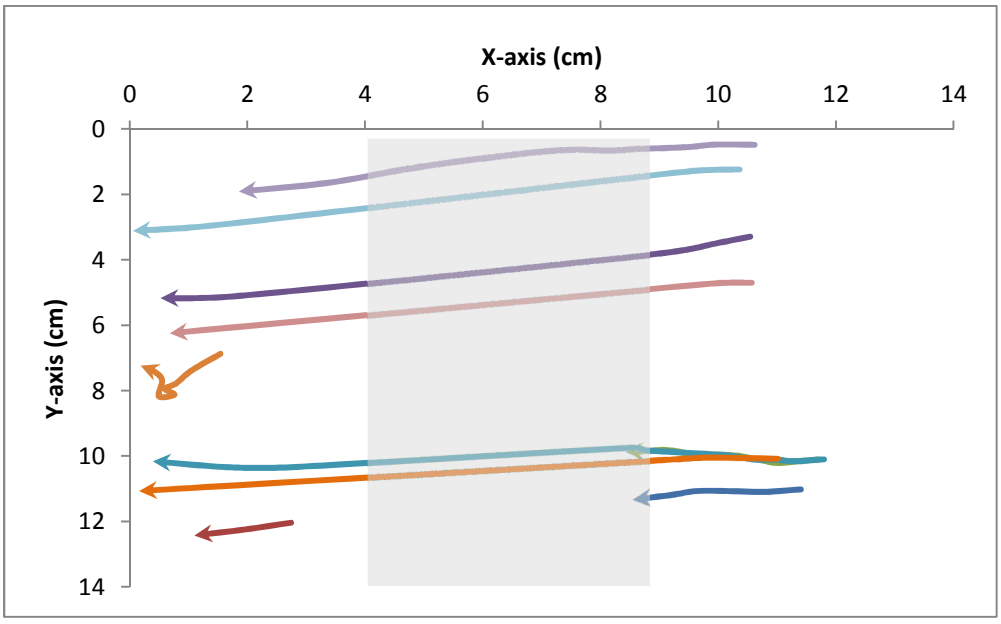


Figure 2.7C: High current speed (31.3 cm s^{-1})



The total number of megalopae counted in the water column increased with current speed, but this trend was not significant (p=0.151), possibly due to recounting of megalopae that exit and re-enter the field of the camera during the lowest current speed (3.5 cm s^{-1}, which is well below the swimming speed of megalopae of 7-12 cm s^{-1}). There was little difference between the number of megalopae in the water column on the highest current speed and the medium current speed.

Tomographic PIV characterization of small scale flows around collector

The small-scale flow patterns were assessed in a 8 x 2 x 4 cm volume directly behind a 5 cm diameter collector under 3 flow velocities (3.5, 20.9, 38.3 cm s^{-1}), and the average vectors were visualized using TechPlot (Figure 2.8). Under the flow conditions tested (3.5 to 38.2 cm s^{-1}), there was an area of lower velocity directly behind the collector that extended about 5 cm downstream of the collector and was below the maximum sustained swimming speed of megalopae reported in the literature of 7-12 cm s^{-1} (Luckenback and Orth 1992). As the current velocity increased, the turbulence behind the collectors increased (measured as turbulent kinetic energy, TKE, Figure 2.9), as did the mean velocity magnitude (Figure 2.10), although an area of "low flow" still existed directly behind the collector. Across the current speeds measured, TKE in this low flow area was still lower than the TKE farther downstream of the collector. When only the x-component of velocity is considered (Figure 2.11), there was some water movement behind the collectors that flows upstream between 3 and 6 cm behind the collector, functioning like an eddy that could carry megalopae back towards the collectors temporarily. This reverse flow is strongest in the mid current speed (20.9 cm s^{-1}), and weaker for the lower (3.5 cm s^{-1}) and higher (38.3 cm s^{-1}) current speeds.

Figure 2.8: Tomographic PIV characterizations of flow behind a scaled down hog's hair collector under low (3.5 cm s^{-1}, A), medium (20 cm s^{-1}, B) and high (38 cm s^{-1}, C) current speeds. Arrows show the vector field across a slice of the field, volumes represent areas of vorticity colored by velocity. Collector is placed upstream just outside of the observation volume (just past -40 mm on X-axis), and water flows in the positive direction along the x-axis.

Figure 2.8A: Lowest current speed (3.5 cm s^{-1})

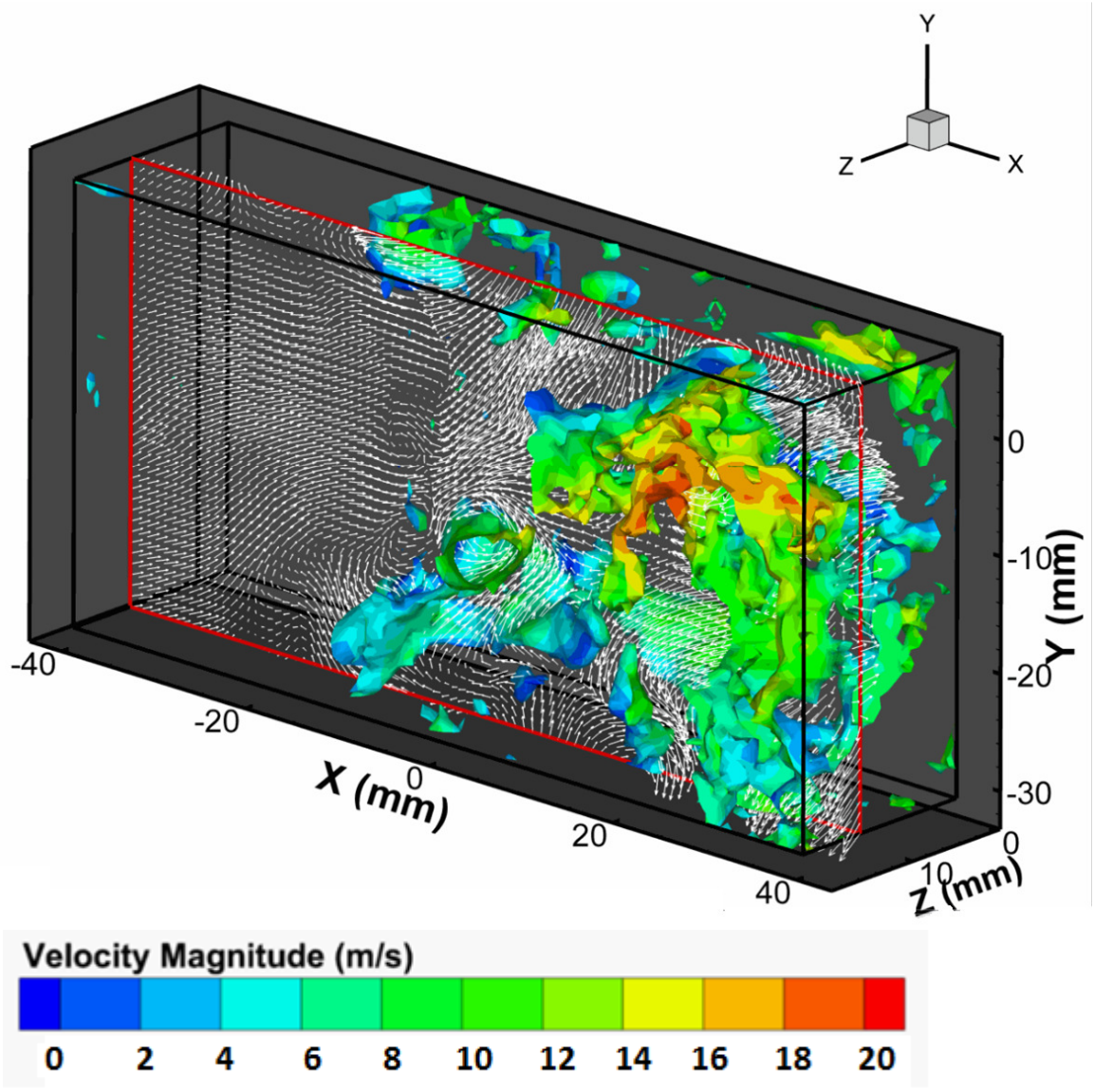
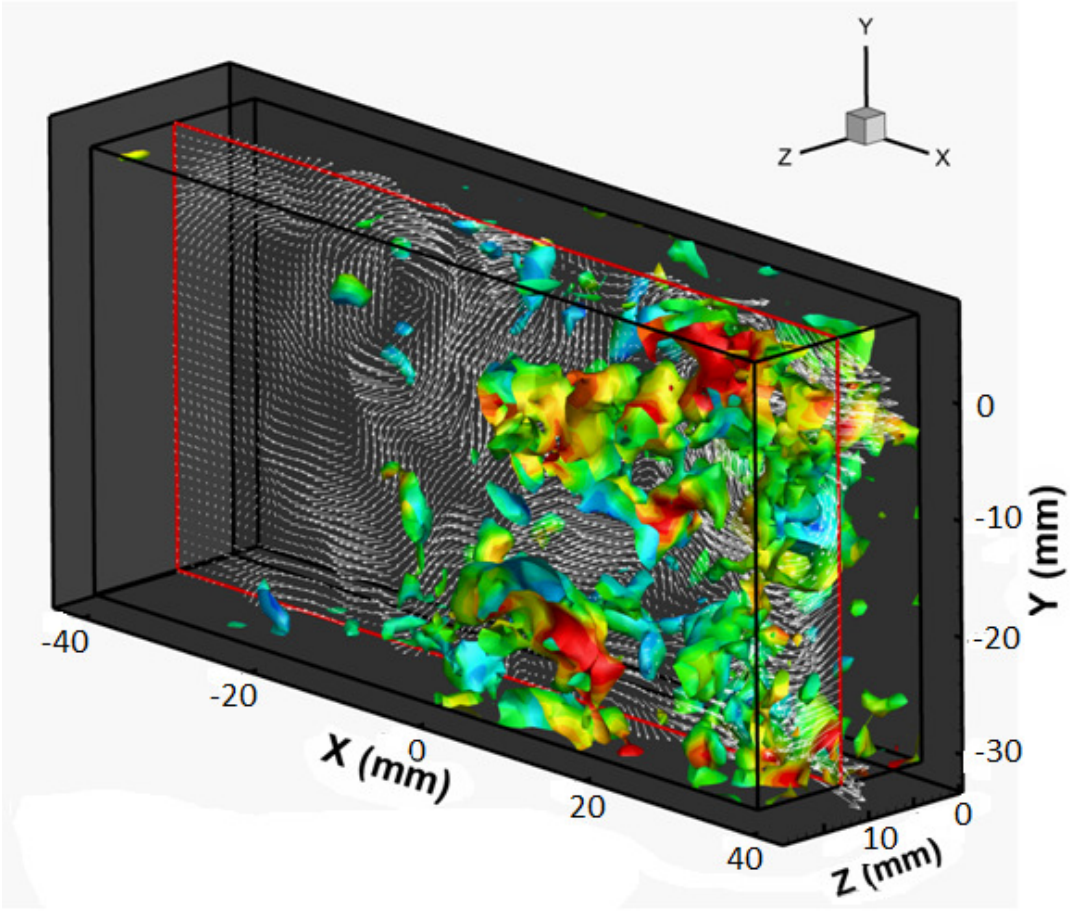


Figure 2.8B: Medium current speed (17.4 cm s^{-1})



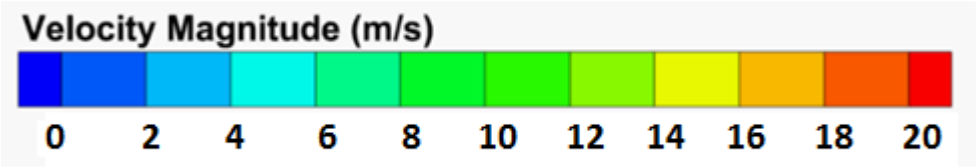


Figure 2.8C: Highest current speed (31.3 cm s^{-1})

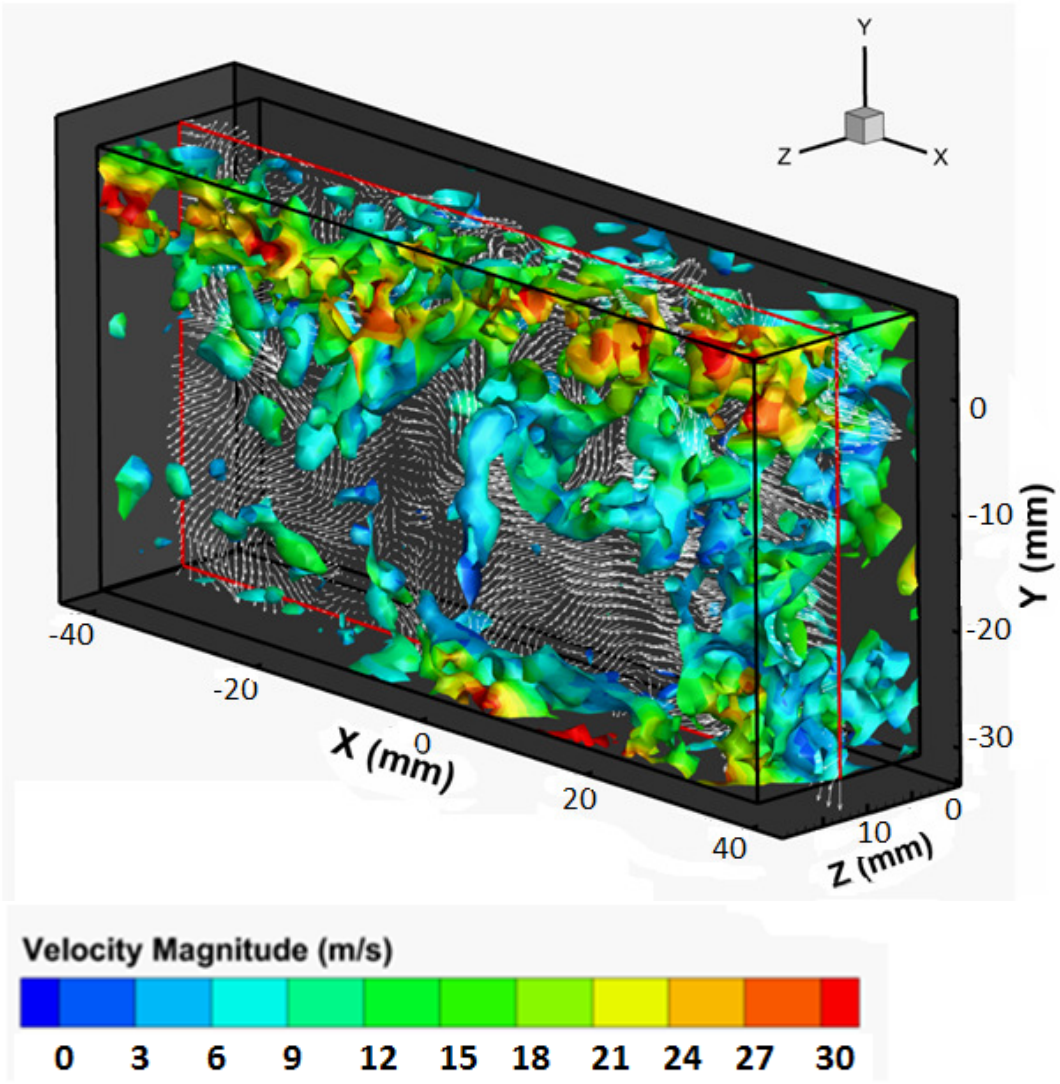


Figure 2.9: Mean Turbulent Kinetic Energy (TKE, $m^2 s^{-2}$) along the X-axis. TKE was calculated for every point in a 1 mm resolution matrix over 10 time steps. TKE was then averaged across the Y and Z planes for visualization along the X-axis. Flow moves in the positive X direction, and the collector is just outside of the measured volume beyond -40 mm X. (rpm1=3.5 cm s^{-1}, rpm5=17.4 cm s^{-1}, rpm9=31.3 cm s^{-1})

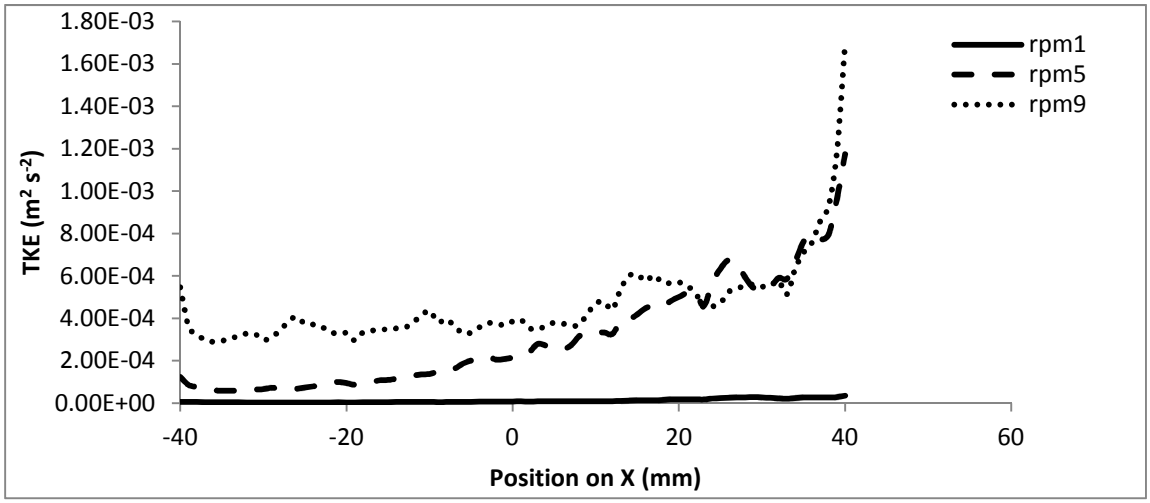


Figure 2.10: Mean velocity magnitude averaged across Y and Z planes and visualized along the X-axis. Flow moves in the positive X direction, and the collector is just outside of the measured volume beyond -40 mm X. (rpm1=3.5 cm s^{-1}, rpm5=17.4 cm s^{-1}, rpm9=31.3 cm s^{-1})

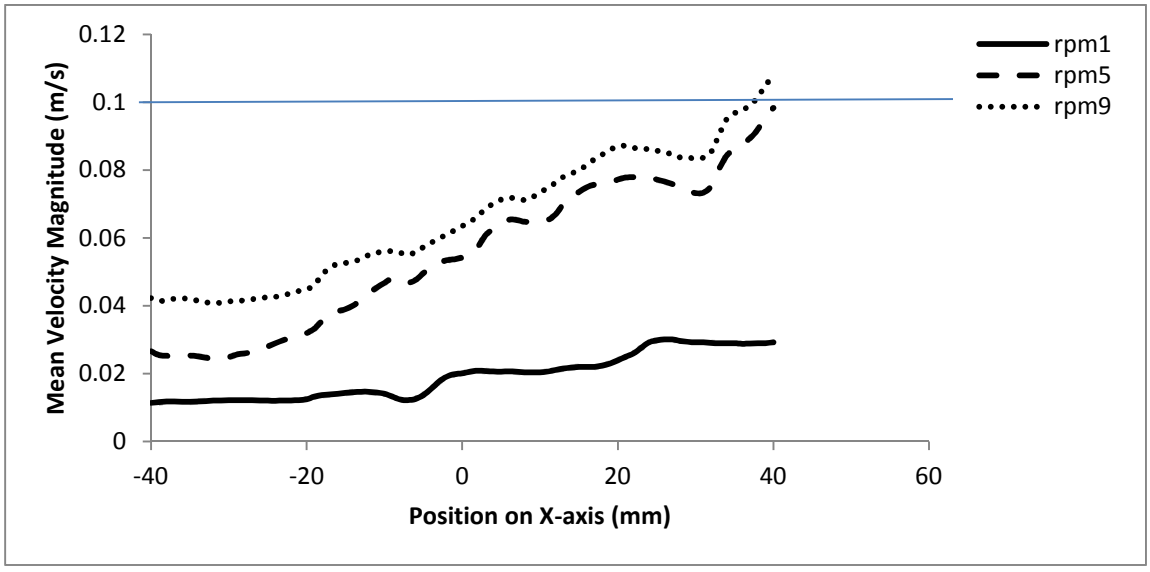
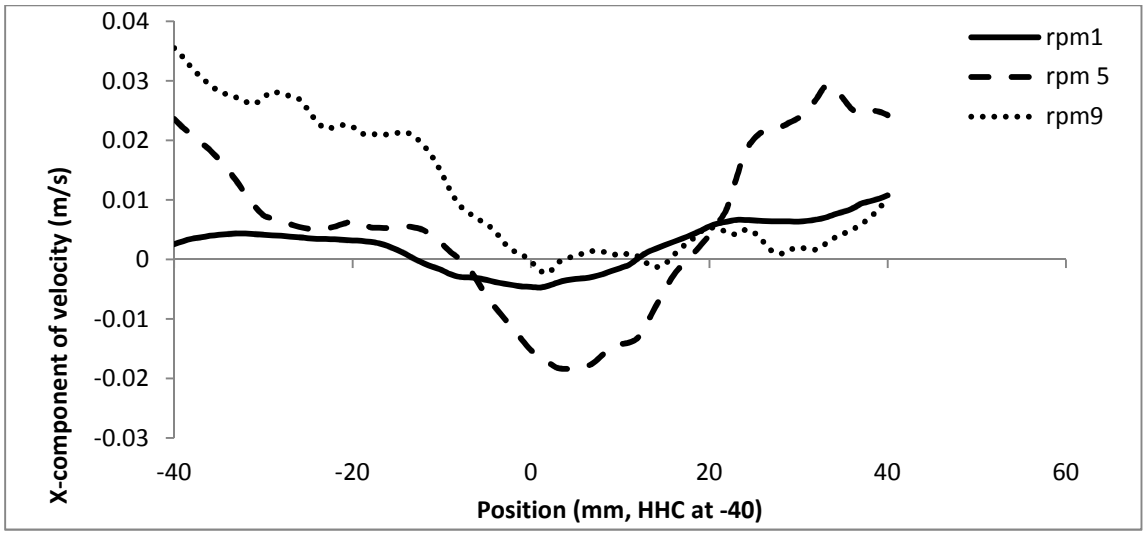


Figure 2.11: Mean X-component of velocity, averaged across Y and Z planes along the X-axis. Negative X-velocities indicate flow moving back towards collector (at -40) and positive X-velocities indicate flow moving away from collector. The collector is just outside of the measured volume beyond -40 mm X. (rpm1=3.5 cm s^{-1}, rpm5=17.4 cm s^{-1}, rpm9=31.3 cm s^{-1})



Encounter rate model

The estimate for R (in Gerritsen and Strickler's 1977 model, or the search radius of megalopae) at slack tide is 21±4 S.E. cm for the large collector and 16±6 S.E. cm for the small collector, which is reasonably within the range that a tiny megalopae could find the relatively large collector, and is within the size range of the collectors (large: 14 cm d. x 25 cm l., small: 5 cm d. x 22 cm d.). Since the collectors are not dimensionless compared to the size of megalopae, we compared the contact surface area (diameter x height) of the collector (large: 0.035 m^2, small: 0.010 m^2) to the surface area of the circle created by the estimate of R (0.138 m^2 large, 0.084 m^2 small). The surface area of the collector is 25% (for large, small: 12%) of the surface area of the circle created by the megalopae's search radius R), indicating that megalopae must be actively sensing and approaching the collector at slack tide rather than physically contacting the collector only

by chance. Adjusting for the surface area of the collector, we re-calculate a distance over which megalopae can sense the collector as 19 cm.

When R was calculated for all time points across a range of velocities (in addition to slack tide), there was a significant exponential decrease in R as "predator speed" (in this case current velocity) increased (Figure 2.12). This relationship fits well to the negative form suggested by Gerritsen and Strickler (1977):

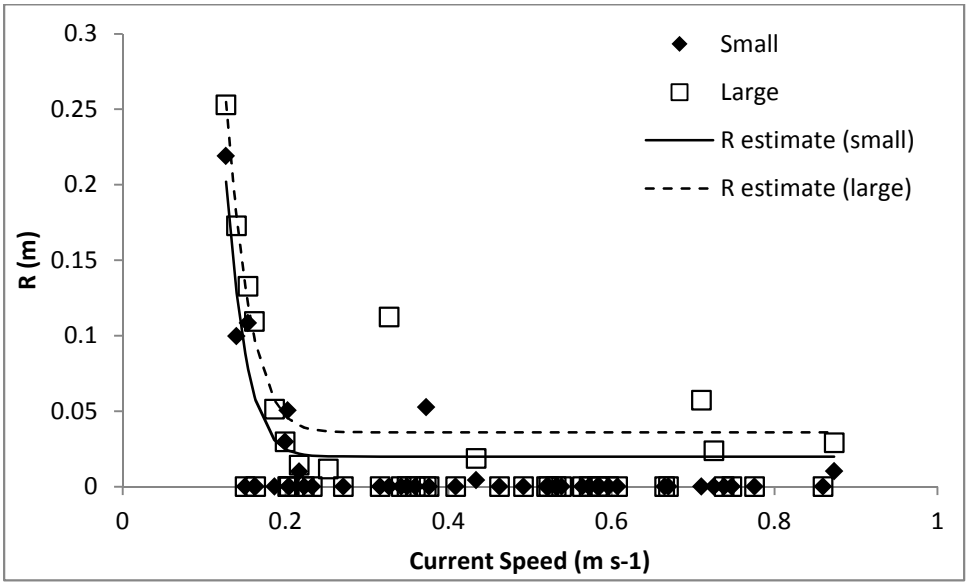
$$R = c_1 * e^{-c_2 v^2} + c_3$$

An iterative linear method was used to parameterize this model for our observed velocities (v) and estimates of R, in which models with the highest r^2 were retained. The model calculated for the large collector (without a plankton net) explained 87.9 % of the variance in R (p=0.0152), and the model of the small collector explained 85.4 % of the variance in R (p=0.017). Although the parameter estimates varied between collector size (Table 2.1), the shapes of the fit models were very similar (Figure 2.12).

Table 2.1: Regression coefficients and fit of search radius (R) to Gerritsen and Strickler's (1977) model for the small and large collectors without plankton nets.

	C_1	C_2	C_3	R^2	p-value
Small	2.07	151	0.02	0.879	<0.001
Large	1.64	125	.036	0.854	<0.001

Figure 2.12: Estimates of R, the search radius of megalopae, as related to collector size and current velocity using the Gerritsen and Strickler (1977) model. R was calculated only with collectors suspended outside of a plankton net, and only the collectors without nets are shown.



DISCUSSION

Hog's hair collectors are widely used, with results published in over 30 peer reviewed papers. However, few studies before this have attempted to interpret the significance of settlement data. The studies that did attempt to compare abundance of megalopae measured in the plankton to settlement generally found poor correlations. Some studies attributed the poor correlations to disparate temporal scales, while others suggested turbulence as a decoupling factor. The design of this study is capable of addressing both potentially decoupling processes. By sampling settlement at precisely the same time frame and spatial location as plankton estimates in the field, we controlled for differences due to spatial or temporal decoupling. By sampling over multiple current speeds in the same night in the field, and through a flume study and tomographic PIV study, we were able to address the second decoupling process of turbulence.

The correlation between abundance of megalopae in the plankton and total number of megalopae settling on collectors deployed at precisely the same time interval and location was strong when current velocity was controlled for (Figure 2.4). Previous studies have reported poor correlations between planktonic abundance of megalopae and settlement (Lipcius et al. 1990, Olmi et al. 1990, Moksnes and Wennhage 2001), even at fine temporal scales (Amaral et al. 2007). The decoupling noted in the other studies may be due to very small scale spatial and temporal variations in the abundance of megalopae or current velocity-based decoupling, both of which were excluded by our study design. Plankton are known to have patchy distributions (Omori and Hamner 1982, Epifanio et al. 1984) that may drive differences between plankton estimates and settlement. Amaral et al. (2007) suggested that turbulence may decouple settlement from supply. We observed a strong exponential negative relationship between the proportion settling on collectors and velocity in the field (Figure 2.5), which was corroborated by a negative velocity coefficient in a regression predicting total settlement based on abundance of megalopae and velocity. Increasing velocity is associated with increasing turbulence, and both support Amaral et al.'s (2007) suggestion that turbulence decouples settlement from the supply of megalopae.

The relationship between settlement and current velocity was also examined in a controlled experimental setting, which found a significant dome shaped relationship between velocity and settlement that predicted maximal settlement within the velocity range of maximal settlement reported in the field (Figure 2.6). While settlement appeared to exclusively rise with decreasing current speed in the field (Figure 2.5), the lowest field velocities measured were significantly higher than the velocities measured during the flume experiment (12 cm s^{-1} minimum in the field, compared to 3.5 cm s^{-1} in the flume).

The lowest current speeds in the field are closer to the mid current speeds tested in the flume, and the strong overlap of the maxima in both the experiment and field suggest that similar processes drive both relationships: (1) a process increasing the abundance of megalopae with increasing velocity (and turbulence) and (2) a process which causes an individual megalopae's ability to settle on a collector to decrease with increasing velocity (and turbulence).

This study provided evidence for both processes driving the dome shaped relationship. In both the field study and the experimental study, the abundance of megalopae in the water column increased with increasing current speed (Figure 2.3). This is consistent with the framework of selective tidal stream transport (Forward et al. 2003) where megalopae keep swimming in response to turbulence. Increasing abundance of megalopae with current speed has also been observed in other field studies (DeVries et al 1994) and laboratory studies (Welch et al. 1999), providing strong support for increasing supply governing the increasing total settlement with current speed up to a maximum speed.

The negative relationship between settlement and velocity at the highest current speed seems to be due to a decrease in contact, as observed in videos of the flume experiment. However, it is unclear if this decrease in contact is due to (1) turbulent cues that signal megalopae to keep swimming, or (2) an inability of megalopae to sense and adhere onto the collector at high speeds. The first mechanism was examined in the small scale measurement of flow and behavioral interactions in the flume study. In the flume experiment, many megalopae approach the collector from downstream (Figure 2.7), where TKE was reduced (Figure 2.9). In the low (3.5 cm s^{-1}) and medium (20.9 cm s^{-1}) current speeds, TKE directly behind the collector was lower than Welch et al.'s (1999)

estimated threshold of 1.1 cm^2 s^2 TKE that stimulates active swimming, although TKE rises back above this value farther downstream in the medium current speed. A decrease in TKE is a strong signal for settlement out of the water column (Welch et al. 1999), so a megalopae passing by the collector that gets drawn into the low TKE area directly behind the collector could sense this cue and settle. At high current speeds (38.3 cm s^{-1}), this low TKE zone is higher than the threshold, but is still reduced compared to areas farther from the collector. Therefore, if megalopae are swept behind the collector, they would receive the correct cue to settle (decrease in TKE).

The predator-prey encounter rate model is a useful framework for examining the second explanation for lowered settlement (inability to sense and settle on the collector at high current speeds) as current speed increases. By comparing estimates of R (the search radius) with current speed, we find that megalopae are less able to sense the collector (and presumably settle) as the current speed increases (Figure 2.12). The exponential decrease in R with increasing speed of the current (which we assign as the "predator speed") is predicted by Gerritsen and Strickler (1977) for plankton interactions. Gerritsen and Strickler (1977) attribute the decrease in R to increasing shear and turbulent forces as velocity increases. The increase in shear and turbulence increases the noise through which a predator (or in our case, the megalopae "prey") must sense prey, and at high levels can override the prey signal, effectively decreasing the search radius. These same forces likely contribute to the decline in settlement with increasing current velocity observed in this study. R is important for understanding the sensory process of settlement, but data from the flume experiment is useful for examining the ability of megalopae to physically settle under variable flume conditions. It is unlikely that megalopae approaching the collector from behind are physically unable to settle, as the

velocity behind the collector at all velocities tested in the flume (up to 38 cm s^{-1}) were consistently below their maximum sustained swimming speed (Figures 2.9, 2.10), but megalopae may be swept past this area of low flow by the current.

The settlement-velocity relationship measured in the flume corresponds to both the encounter rate based model of settlement and the empirically derived linear relationship between velocity, abundance, and settlement in the field. While settlement in the flume was low at the lowest velocities tested, and highest at the lowest velocities measured in the field, the lowest flume speed (3 cm s^{-1}) was far below any current speed measured in the field (10 cm s^{-1}). The maxima of both settlement curves are in the 15-20 cm s^{-1} range, which corresponds to the minimum velocities that were observed at slack tide in the field. Our result of highest settlement at slack tide is consistent with a previous study in North Carolina (Tankersley et al. 1997).

To generalize our approach and aid interpretation of settlement data at other sites, we can re-frame settlement using the encounter-rate model of Gerritsen and Strickler (1977). Since R (search radius) is likely a sensory process that does not rely on light (megalopae in water column during night) or chemical (artificial settlement substrates have no scent) sensory processes, R is likely to be consistent between sites. In this case, settlement can be generalized as:

$$Settlement = \int \pi * R^2 * N * \frac{v_{megalopae}^2 + 3v_{current}^2}{3v_{current}}\, dt$$

We replace the *R* term with our velocity-based estimate for the large collectors (Table 2.1), N with the empirically derived relationship between plankton abundance and velocity (Figure 2.3), and $v_{megalopae}$ as 0.1 m s^{-1} (Olmi 1992). The resulting formula relies

only on current velocity and the nightly intercept of the abundance-velocity model (α_i) to predict settlement:

$$Settlement = \int \pi * (9.85e^{-130v^2} + 0.043)^2 * (e^{\propto_i + 1.14 * v}) * \frac{0.1^2 + 3v^2}{3v}\, dt$$

In practice, the estimates of *v* will not be a continuous function but a series of discrete measurements taken at a time interval *dt*. Thus, settlement can be framed as a sum of terms:

$$Settlement = \sum_{n=0}^{n=k} \pi * \left(c_1 e^{-c_2 v_n^2} + c_3\right)^2 * (e^{\propto_i + 1.14 * v_n}) * \frac{0.1^2 + 3v_n^2}{3v_n} dt$$

Where n=0 is the first time point after dark, and n=k is the last time point before sunrise. Since megalopae are generally only in the water column by night (Forward and Rittschof 1994), considering only nighttime values avoids underestimating the true abundance based on daily measures of settlement. The velocity-independent abundance metric, α_i, can be calculated by re-arranging the summation above:

$$\alpha_i = \ln\left(\frac{Settlement}{\sum_{n=0}^{n=k} \pi * \left(c_1 e^{-c_2 v_n^2} + c_3\right)^2 * \frac{0.1^2 + 3v_n^2}{3v_n} * dt * e^{1.14 * v_n}}\right)$$

The α_i term alters the total settlement measured during the course of a night, but not the relationship between velocity and settlement. Thus, the velocity of maximal settlement is unaltered between nights. The predicted maximum settlement is at the lowest current speeds, and consistent with the maximum settlement at slack tide in the field. The term α_i is velocity-independent, and is therefore comparable between nights at

the same site that experienced different flow conditions (as is often the case in semi-diurnal mixed tidal regimes of the Gulf of Mexico). Once α_i is known, abundance can be compared between nights without the bias or decoupling effects of current velocity, and flux can be estimated if recruitment is a research question of interest:

$$flux = \sum_{n=0}^{n=k} e^{\alpha_i + 1.14 * v} dt$$

Caution should be taken when attempting to estimate flux of megalopae into the estuary, as differences in transport on the ebb and flood tide are likely and cannot be accounted for by collectors deployed over both tidal cycles throughout the night. Megalopae were caught in high abundance on the ebb tide during this study, making estimates of net flux on given nights difficult to disentangle

It is important to note that formulas estimating the abundance of megalopae in relation to current speed may vary between sites, and should be empirically validated before applying this approach to new sites. Additionally, the fit of R in relation to current speed may vary based on the shape and size of collector, and should also be validated before application to a new system. However, both relationships can easily be estimated with the approaches described in this paper.

Overall, this study has shown that settlement on hog's hair collectors is a reasonable measure of the supply of megalopae at a given site when current velocity is accounted for. Although turbulence decouples settlement from abundance, the decoupling effect can be controlled for by controlling for current velocity. Turbulence and current speed have a high impact on the search radius of megalopae, and decouple the abundance of megalopae from settlement when compared between high and low current speeds. Comparisons of settlement between sites that have different hydrology characteristics are

unwise, as are comparisons within sites that were made between nights with different flow conditions. However, comparisons between nights and potentially sites are possible when velocity data is available, and relationships between velocity, abundance of megalopae in the plankton, and settlement can be estimated.

References

Adhikari, D, and E Longmire (2013) Infrared tomographic PIV and 3D motion tracking system applied to aquatic predator-prey interaction. Meas. Sci. and Tech. 24: 024011.

Aguilar, R, AH Hines, TG Wolcott, DL Wolcott, MA Kramer, RN Lipcius (2005) The timing and route of movement and migration of post-copulatory female blue crabs, *Callinectes sapidus* Rathbun, from the upper Chesapeake Bay. J Exp Mar Bio Eco. 319, 117-128.

Amaral, V, H Queiroga, M Skov, J Paula (2007) Planktonic availability and settlement of *Carcinus maenas* megalopae at high temporal resolution in the lower Mira Estuary (SW Portugal). Mar Ecol Prog Ser 348:239-248.

Brown, CA, SA Holt, GA Jackson, DA Brooks, GJ Holt (2004) Simulating larval supply to estuarine nursery areas: how important are physical processes to the supply of larvae to the Aransas Pass inlet? Fish Ocean 13:181-196.

Brown, CA, GA Jackson, SA Holt, GJ Holt (2005) Spatial and temporal patterns in modeled particle transport to estuarine habitat with comparisons to larval fish settlement patterns. Estuarine, Coastal, and Shelf Science, 64:33-46

Carr, SD, RA Tankersley, JL Hench, RB Forward Jr, RA Luettich Jr (2004). Movement patterns and trajectories of ovigerous blue crabs *Callinectes sapidus* during the spawning migration. Estuarine, Coastal and Shelf Science 60, 567–579.

CDMO (2013). Central Data Management cdmo.baruch.sc.edu

Chavez-Ramirez F (1996) Food availability, foraging ecology,and energetics of whooping cranes wintering in Texas. PhD dissertation, Texas A&M University, College Station

Colin, SP, HG Dam (2002). Latitudinal differentiation in the effects of the toxic dinoflagellate *Alexandrium* spp. on the feeding and reproduction of populations of the copepod *Acartia hudsonica*. Harmful Algae 1, 113–125.

Cronin TW, RB Forward (1988) The visual pigments of crabs I. Spectral characteristics. J Comp Physiol A 162: 463-478

Daewel, U, C Schrum, A Temming (2011). Towards a more complete understanding of the life cycle of brown shrimp (*Crangon crangon*): modelling passive larvae and juvenile transport in combination with physically forced vertical juvenile migration. Fisheries Oceanography 20, 479–496.

DeVries, MC, RA Tankersley, RB Forward, WW Kirby-Smith, RA Luettich (1994) Abundance of estuarine crab larvae is associated with tidal hydrologic variables. Mar Biol 118:403-413.

Epifanio, CE, CC Valenti, AE Pembroke (1984) Dispersal and recruitment of blue crab larvae in Delaware Bay, USA. Est Coast Shelf Sci 18.1:1-12.

Etherington LL, DB Eggleston (2000) Large-scale blue crab recruitment: linking postlarval transport, post-settlement planktonic dispersal, and multiple nursery habitats. Mar Ecol Prog Ser 204:179-198.

Flores, AV, J Cruz, J Paula (2002) Temporal and spatial patterns of settlement of brachyuran crab megalopae at a rocky coast in Central Portugal. Mar Eco Prog Ser 229:207-220.

Forward, RB, JH Coen, RD Irvine, JL Lax, R Mitchell, AM Schinck, MM Smith, JM Thompson, JI Venezia (2004) Settlement of blue crab *Callinectes sapidus* megalopae in a North Carolina estuary. Mar Ecol Prog Ser 269: 237-247.

Forward RB, TW Cronin, DE Stearns (1984) Control of diel vertical migration: photoresponses of a larval crustacean. Limn and Ocean 29: 146-154.

Forward RB, D Rittschof (1994) Photoresponses of crab megalopae in offshore and estuarine waters: Implications for transport. J Exp Mar Bio and Eco 182: 183-192.

Forward RB, J Swanson, RA Tankersley, JM Welch (1997) Endogenous swimming rhythms of blue crab, *Callinectes sapidus*, megalopae: effects of offshore and estuarine cues. Mar Biol 127:621-628.

Forward RB, J Swanson, RA Tankersley, JM Welch (2003) Selective tidal-stream transport of the blue crab *Callinectes sapidus*: an overview. Bul Mar Sci 72(2): 347-365.

Forward RB, RA Tankersley, D Blondel, D Rittschof (1997) Metamorphosis of the blue crab *Callinectes sapidus*: effects of humic acids and ammonium. Mar Ecol Pro Ser 157:277-286.

Forward RB, RA Tankersley, D Rittschof (2001) Cues for metamorphosis of brachyuran crabs: an overview. Amer Zool 41:1108-1122.

Forward RB, RA Tankersley, KA Smith, JM Welch (2003) Effects of chemical cues on orientation of blue crab, *Callinectes sapidus*, megalopae in flow: implications for location of nursery areas. Mar Biol 142: 747-756.

Forward, RB, RA Tankersley, JM Welch (2003) Selective tidal-stream transport of the blue crab *Callinectes sapidus*: an overview. Bul. Mar. Sci 72: 347-365.

Forward, RB, MC De Vries, D Rittschof, DA Frankel, JP Bischoff, CM Fisher, JM Welch (1996) Effects of environmental cues on metamorphosis of the blue crab *Callinectes sapidus*. Mar Ecol Prog Ser 131: 165-177.

Gerritsen J, JR Strickler (1977) Encounter probabilities and community structure in zooplankton: a mathematical model. J. Fish. Bd. Can 31(1): 73-82.

Goodrich, DM, J van Montfrans, RJ Orth (1989) Blue crab megalopal influx to Chesapeake Bay: Evidence for a wind-driven mechanism. Est Coas Shelf Sci 29:247-260.

Guillory, V (2000) Relationship of blue crab abundance to river discharge and salinity. Proc Annu Conf SEAFWA 54:213-220.

Hasek, BE, NN Rabalais (2001) A comparison of molt states of blue crab megalopae, *Callinectes sapidus* (Rathburn), sampled with artificial collectors and plankton nets. J Exp Mar Biol and Eco 265:15-27.

Heck, KL, L Coen (1995) Predation and the abundance of juvenile blue crabs: a comparison of selected east and gulf coast (USA) studies. Bull Mar Sci 57: 877-883.

Hoeinghaus DJ, SE Davis (2007) Size-based trophic shifts of saltmarsh dwelling blue crabs elucidated by dual stable C and N isotope analyses. Mar Ecol Pro Ser 334: 199-204.

Johnson, DR, HM Perry (1999). Blue crab larval dispersion and retention in the Mississippi Bight. Bulletin of Marine Science 65, 129–149.

Kim, IN. (2009) Analysis of physical mixing processes between estuaries and the Gulf of Mexico at the Aransas Pass tidal inlet, TX. Final Report to Mission-Aransas National Estuarine Research Reserve Graduate Research Assistant FY2008.

Kingsford, MJ, JM Leis, A Shanks, KC Lindeman, SG Morgan, J Pineda (2002). Sensory environments, larval abilities and local self-recruitment. Bulletin of Marine Science 70, 309–340.

Kordos, LM, RS Burton (1993). Genetic differentiation of Texas Gulf Coast populations of the blue crab *Callinectes sapidus*. Marine Biology 117, 227–233.

Lipcius, RN, EJ Olmi, J van Montfrans (1990) Planktonic availability, molt stage and settlement of blue crab postlarvae. Mar Eco Pro Ser 58:235-242.

Longely WL (1994) Freshwater inflows to texas bays and estuaries: ecological relationships and methods for determination of needs. Texas Water Development Board, Austin, TX.

Luckenbach, MW, RJ Orth (1992) Swimming velocities and behavior of blue crab (*Callinectes sapidus* Rathburn) megalopae in still and flowing water. Ests 15.2: 186-192.

MacKenzie, BR, TJ Miller, S Cyr, WC Leggett (1994) Evidence for a dome shaped relationship between turbulence and larval fish ingestion rates. Limnol. Oceanogr. 39(8): 1790-1799.

McMillen-Jackson, AL, TM Bert, P Steele (1994). Population genetics of the blue crab *Callinectes sapidus*: modest population structuring in a background of high gene flow. Marine Biology 118, 53–65.

Mense, DJ, EL Wenner (1989) Distribution and abundance of early life history stages of the blue crab, *Callinectes sapidus*, in tidal marsh creeks near Charleston, South Carolina. Ests. 12(3): 157-168.

Metcalf, KS, RN Lipcius (1992) Relationship of habitat and spatial scale physiological state and settlement of blue crab postlarvae in Chesapeake Bay. Mar Ecol Prog Ser 82: 143-150.

Minello TJ, KW Able, MP Weinstein, CG Hays (2003) Salt marshes as nurseries for nekton: testing hypotheses on density, growth and survival through meta-analysis. Mar Ecol Prog Ser 246:39-59.

Moksnes, P, H Wennhage (2001) Methods for estimating decapods larval supply and settlement: importance of larval behavior and development stage. Mar Eco Prog Ser 209:257-273.

Montagna, P, J Brenner, J Gibeaut, S Morehead (2011) Chapter 4: Coastal Impacts. In: The Impact of Global Warming on Texas. Pub. Texas Climate Initiative, *eds.* J. Schmandt, J. Clarkson, G.R. North.

More W (1969) A contribution to the biology of the blue crab in Texas, with a description of the fishery. TPWD Technical series 1: 1-31.

Morgan, SG, RK Zimmer-Faust, KL Heck, LD Coen (1996) Population regulation of blue crabs *Callinectes sapidus* in the northern Gulf of Mexico: postlarval supply. Mar Ecol Prog Ser 133:73-88.

NOAA (2006) Final Programmatic environmental impact statement: federal approval of the Texas national estuarine research reserve and management plan, the Mission-Aransas Estuary. U.S. Dept. Commerce, NOAA, NOS, Silver Spring, MD.

North, EW, Z Schlag, RR Hood, M Li, L Zhong, T Gross, VS Kennedy (2008). Vertical swimming behavior influences the dispersal of simulated oyster larvae in a coupled particle-tracking and hydrodynamic model of Chesapeake Bay. Mar Ecol Prog Sers: 359, 99.

Ogburn, MB, H Diaz, RB Forward (2009) Mechanisms regulating estuarine ingress of blue crab *Callinectes sapidus* megalopae. Mar Eco Prog Ser 389: 181-192.

Olmi, EJ, J van Montfrans, RN Lipcius, RJ Orth, PW Sadler (1990) Variation in planktonic availability and settlement of blue crab megalopae in the York River, Virginia. Bul Mar Sci 46:230-243.

Omori, M, WM Hamner (1982) Patchy distribution of zooplankton: behavior, population assessment and sampling problems. Mar. Biol. 72: 193-200.

Paula, J, C Bartilotti, T Dray, A Macia, H Queiroga (2004). Patterns of temporal occurrence of brachyuran crab larvae at Saco mangrove creek, Inhaca Island (South Mozambique): implications for flux and recruitment. Journal of Plankton Research 26, 1163–1174.

Posey, MH, TD Alphin, H Harwell, B Allen (2005) Importance of low salinity areas for juvenile blue crabs, *Callinectus sapidus* Rathburn, in river-dominated estuaries of south eastern United States. J Exp Mar Biol and Eco 319: 81-100.

Powell, G, J Matsumoto, D Brouck (2002) Methods for determining minimum freshwater inflow needs for texas bays and estuaries. Estuaries 25: 1262-1274.

R Core Team (2013) R: a language and environment for statistical computing. R Foundation for statistical computing, Vienna, Austria. http://www.R-project.org/

Rabalais NN, FR Burditt, LD Coen, BE Cole, C Eleuterius, KL Heck, TA McTigue, SG Morgan, HM Perry, FM Truesdale, RK Simmer-Faust and RJ Zimmerman (1995) Settlement of *Callinectes sapidus* megalopae on artificial collectors in four gulf of mexico estuaries. Bul Mar Sci 57(3): 855-876.

Reese, MM, GW Stunz, AM Bushon (2008) Recruitment of estuarine-dependent nekton through a new tidal inlet: the opening of Packery Channel in Coprus Christi, TX, USA. Est and Coasts 31:1143-1157.

Roman, MR, and WC Boicourt (1999). Dispersion and recruitment of crab larvae in the Chesapeake Bay plume: physical and biological controls. Estuaries 22, 563–574.

Scharf FS, KK Schlicht (2000) Feeding habits of red drum in Galveston Bay, Texas: seasonal diet variation and predator-prey size relationships. Est and Coasts 23: 128-139.

Smith, NP (1977) Meterological and tidal exchanges between Corpus Christi Bay, Texas, and the Northwestern Gulf of Mexico. Est. Coast. Mar. Sci. 5: 511-520.

Tankersley RA, LM McKelvey, RB Forward (1995) Responses of estuarine crab megalopae to pressure, salinity and light: implications for flood-tide transport. Mar Biol 122: 391-400.

Tankersley, RA, JM Welch, RB Forward (2002) Settlement times of blue crab (*Callinectes sapidus*) megalopae during flood-tide transport. Mar Biol 141:863-875.

Tankersley, RA, RB Forward (1994) Endogenous swimming rhythms in estuarine crab megalopae: implications for flood-tide transport. Mar Bio 118: 415-423.

Taylor, AC, E Naylor (1977). Entrainment of the locomotor rhythm of *Carcinus* by cycles of salinity change. J. Mar. Biol. Assoc. UK 57, 273–277.

Thacker WC (2007) Estimating salinity to complement observed temperature: 1. Gulf of Mexico. J Mar Syst 65:224-228.

Welch JM, RB Forward (2001) Flood tide transport of blue crab, *Callinectes sapidus,* postlarvae: behavioral responses to salinity and turbulence. Mar Biol 139: 911-918.

Welch JM, RB Forward, PA Howd (1999) Behavioral responses of blue crab *Callinectes sapidus* postlarvae to turbulence: implications for selective tidal stream transport. Mar Ecol Prog Ser 179: 135-143.

Welch JM, D Rittschof, TM Bullock, RB Forward (1997) Effects of chemical cues on settlement behavior of blue crab *Callinectes sapidus* postlarvae. Mar Ecol Prog Ser 154: 143-153.

Wilber DH (1993) The influence of Apalachicola River flows on blue crab, *Callinectes sapidus*, in north Florida. Fish Bul 92:180-188.

Vita

Kimberly Bittler is originally from Maryland, completing her Bachelor's Degree at Colby College in Maine before coming to the University of Texas for her Master's Degree in Marine Science.

Permanent email: kmbittler@gmail.com

This thesis was typed by Kimberly Bittler.